STUDIES ON GROWTH AND EFFICIENCY OF FOOD UTILIZATION IN RATS.

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CONTENTS.

PART I

THE EFFECTS OF GONADECTOMY ON FOOD UTILIZATION AND THE GROWTH OF BODY AND ORGANS IN RATS.

Castration has been practiced on male domestic animals from time immemorial. There are probably a variety of reasons for this, among which greater ease of management, reduced activity and faster fattening are doubtless of importance. Further, the quality of the meat in respect of both texture and flavour is improved. The spaying of females, particularly among pigs, was also a common practice which has now almost disappeared. Since spaying is a risky operation under farmyard conditions , there must have been a strong belief that the advantages were real and considerable. These advantages appear to have been the absence of heat periods affecting growth rate and a supposed greater economy of food consumption for a given weight increase. It is a common belief that castrated males and spayed females convert food-stuffs into edible meat more quickly than they would otherwise, and hence the impression may have arisen that they were at the same time more economical in the use of food. So far, however, there have been few experiments on either farm or laboratory animals under strictly controlled environmental conditions to support this view. The possibility has to be borne in mind that an increased rate of gain may not necessarily be accompanied by improved economy in food utilization. Unless the food consumption is taken into consideration, there is no justification for assuming that faster growth rate means greater economy of gain. The centuries-long experience of farmers may serve as evidence that the rate of gain in body weight is improved by castration, but to a scientific agriculturist it would be interesting and useful to know approximately how much can be effected by this operation on different kinds of animals and at different ages. In recent years, research workers in the field of nutrition have done much to investigate the efficiency of food utilization as influenced by different quantities as well as qualities available for growth and maintenance, yet very few have centered their interest on the effects of castration.

In female domesticated animals spaying is not so widely applied as the castration of males. This fact may be attributed to the general view that energy loss due to sexual excitement is less pronounced than in males, and also that the quality of the meat does not seem to be much affected by the presence of ovaries. However, should the spayed animals prove to grow faster or more economically than the unspayed and are otherwise suitable the practice might be extended to them in commercial feeding.

With the advancement of knowledge of endocrine functions the interdependence of hormones from different glands in controlling the functioning and well-being of animal life is being more fully recognised. Either excessive or deficient secretion from one of the endocrine glands will result in a disturbance of physiological activities and changes in the normal growth curve of the body and in the development of various organs. The gonads are not solely concerned with reproduction of the animal - they also assert a strong influence on metabolism and growth as well. Gonadectomy eliminates the secretion of hormones from one of the principle endocrine organs, so that the changes taking place after the operation are of considerable interest to physiologists. A certain amount of work has been reported on the effect of gonadectomy on the growth of body and organs, but the results are in many cases contradictory, especially in respect of castrated males. Further investigation of this matter is therefore indicated. It has a direct practical importance because apart from the question of economics involved in efficiency of food utilization, it has become essential to discover the factors underlying quality in meat products. One of these factors is the carcass proportions of bone, muscle and fat. It is known that the relative rates of growth of the body and its component parts undergo changes with age, and these changes are not the same in the two sexes. It is desirable to know therefore what differences in the proportions of the various organs and tissues may be traced to the endocrine system of the animals. If the rate of growth can be affected by the presence or absence of the gonads, it is clearly of interest to determine the time at which the gonads exert their effects. It may be that the time of castration might have some influence on the carcass quality by altering the relative rates of growth of bone and fat at various ages of the animal.

With these considerations in mind experiments with rats have been carried out with the object of observing the changes taking place in the rate and economy of gain in rats of both sexes following gonadectomy at various ages. An attempt has also been made to relate these changes to the sizes of the organs and tissues making up the total body weight. It is realized that the direct transference of conclusions from such studies as these to problems of growth and carcass quality in farm animals would be unwise, but if the results suggest profitable types of experiment with larger animals the purpose of the present experiments is well served.

REVIEW OF LITERATURE.

Early investigations on the effect of castration were chiefly concerned with the development of secondary sexual characteristics modified by the removal of sex glands. At the beginning of the present century physiologists began to be interested in the relationship between gonads and other organs of internal secretion, and some observations were made on the growth of endocrine glands and growth of skeleton in castrated animals. However, these experiments, especially in larger animals, were carried out on small numbers and with inadequate controls, and the animals used might have been of different ages, litters, or even strains, and were certainly not always comparable. A systematic study on the growth of laboratory animals after castration was for the first time reported in 1909 by Stotsenburg on albino rats. His repert was based on the data collected from three series of experiments running during the years from 1905 to 1908, with 99 animals, including 52 castrated and 47 controls. He concluded that in the case of albino rats, the growth curve for the castrated is similar to that for the normals. It is true, as indicated from the tables and graphs he presented, that the growth curves of the male rats with and without gonads are almost identical, yet we should also notice that in all three series the final weight of castrates is lighter, which may still be within the normal fluctuations, but this tendency is consistent. Moreover, if the tables of body weight at different ages in his report are examined it will be seen that the number of animals he averaged was not more than half of the total which he used in the experiment, and not infrequently he only took one third or one fourth, even as few as one tenth of the animals. He intimated in the tables that observations on some litters were discontinued due to illness, but he did not give reasons for others who were apparently not subject to diseases.

In regard to the growth of female animals after ovariectomy, Stotsenburg (1913) was also the first investigator. He found that the average body weight of spayed rats in three series (1909 to 1911) ranged from 17.1 per cent to 30 per cent greater than control females. This was attributed to general overgrowth or to the deposition of fat, or to both of these causes.

Hatai (1913) made postmortem examinations on the materials used by Stotsenburg to determine the effect of gonadectomy on the weight of the central nervous system and of the hypophysis, and reported that there was a striking increase in the weight of hypophysis in castrated rats, but practically no change after spaying. He confirmed Stotsenburg's findings on the growth of body weight. Hatai recognised that the direct comparison of observed values is not always justifiable, so he devised formulae for a more exact comparison. In a later publication, Hatai (1915) carried out an extensive investigation on the effect of gonadectomy on the growth of the body and endocrine glands. This study comprises five different operations; namely, (1) total gonadectomy -- castration and spaying;

(2) partial gonadectomy -- semi-castration and semi-spaying; (3) ligation of spermatic cord; (4) removal of one ovary, followed by an isolation of the other ovary from the uterus; and (5) the isolation of both ovaries from the uterus. So far as the growth of body is concerned, his results substantiated his previous findings.

Bugbee (1926) observed a higher growth rate in gonadectomised male and female rats. He used only six castrated rats, and three of them were injected with ovarian folliculin hormone, so that the evidence seems inadequate.

van Wagenen (1928) reported that male albino rats castrated at the time of weaning do not maintain the body weight and body length of the controls. The mean weight of the castrated rats in one series was 25.8 per cent less than their controls at 350 days of age, and in another series the difference at 450 days age was 20.7 per cent in favour of the controls. He stated that this failure was not always evident during the first 100 to 150 days, but thereafter there was an increasing divergence between curves for body weight and length growth of tested and controls. The tail length does not respond to castration consistently, therefore he asserted that body length rather than total length is the significant linear measurement.

Freudenberger and co-workers (Freudenberger and B illeter 1935; Freudenberger and Howard 1937; Freudenberger and Hashimoto 1937) Studied the effect of ovariectomy on the growth of body and organs, and concluded that the spayed albino rats are significantly greater in body weight than the controls.

Commins (1932), Korenchevsky (1934) and Lawless (1936) all reported decreased body weight in castrated rats. Moore (1922) observed smaller growth of body weight in gonadectomised guinea pigs. Working with mice, Masui and Tamura (1925) were unable to find out any change in body weight growth in castrated males, but the spayed females grew more rapidly.

Slonaker (1930) investigated the effect of gonadectomy in rats in connection with food intake. Besides he also studied the spontaneous running activity, growth and life span. He found, from the eight castrated male rats and thirty-seven spayed females used, that the gonadectomised animals had the greatest maximum weight in both sexes. The food consumed by normal males is almost twice as much as that consumed by castrates, while normal females consumed 39.33 per cent more than spayed ones. The average daily running activity in number of revolutions (as recorded in revolving cages) at the age of 200 days for normal males was 12798, and for castrated was 179, being only 0.014 per cent. In females, it was 11068 for normals, but only 123 for spayed. The life span of gonadectomised rats is shorter than that of their respective controls.

Holt, Keeton and Vennesland (1936) used 54 rats in their experiment, comprising 26 males and 28 females, one half of each were gonadectomised and the rest were kept as controls. The animals were operated between 33 and 73 days of age. They found that all spayed females had greater body weight during the course of fifty-two weeks. In two out of their four litters, the male controls gained and maintained greater weight than castrated, yet in the other two litters the castrates exceeded controls in growth. Therefore they concluded "the weight of males following castration varied. The variations are not predictable and are not significant." There was no significant difference in the food intake of castrated and control males. Although the actual food consumption of spayed females was greater than that of of their controls, yet relative to body weight it was somewhat less.

The most recent paper seen is that of Sanderg, Perla and Holly (1939) who studied the effects of excission of various endocrine glands on metabolism, recording the changes of body weight and food intake. They gonadectomised eighteen albino rats before puberty (7 weeks of age) and eighteen rats after puberty (6 months old). They found that the growth curves of normal and castrated males were similar up to the age of 30 weeks. Thereafter the curve of the castrates flattened out, and at the age of 40 weeks the intact animals weighed 10 per cent more. Food consumption, expressed in grams per 100 grams of body weight, was somewhat higher in the controls up to a weight of 270 grams. After that both groups ate practically the same amount of food. Female rats spayed before puberty showed a weight curve similar to that of the normal group. They consumed the same amount of food per 100 grams of body weight as controls. Females spayed after puberty showed a definite gain in weight over controls up to the end of the experiment (43 weeks of age). This gain was acheived in spite of the fact that their food intake, expressed in grams per 100 grams of body weight, was less than that of the controls. Since they did not give exact figures or graphs of body weight and food consumption, it is difficult to judge the value of this work. In the report they only stated that they had gonadectomised thirty-six rats, without mentioning the numbers in each sex. Further, with regard to controls, they said that the "young" rats (operated before puberty) were studied for a control period of four weeks before castration and for seven months afterwards. The "adult" rats (operated after puberty) were observed for a control period of four weeks before removal of the gonads and four months afterwards. It remains doubtful on what bases their comparison of growth and food intake were actually made. Furthermore, they appear to regard the albino rats of 7 weeks of age as not having reached puberty, yet reports(Hain 1934) intimate that females reach sexual maturity at 40 to 45 days of age.

Reports on the effect of gonadectomy on the modification of secondary sexual characteristics such as horn and feather growth in farm livestock and fowls are abundant. However, as far as growth of body or efficiency of food utilization is concerned, investigations are few and rather inadequate, owing no doubt to the technical difficulties of carrying them out. A comparatively extensive investigation was made by MacKenzie and Marshall (1915) who compared the growth and carcass weights of 30 spayed gilts with 30 control litter-mates. In spite of considerable variation, the spayed gilts were considered superior in growth rate (A statistical test made on the published data shows that the authors were justified, the probability that the observed difference was a chance one being only about 2 per cent). In this experiment food consumptions of individual pigs were not recorded so that nothing could be said about the efficiency of the two groups of pigs.

Stahl (1934) arrived at a different conclusion in regard to the effect of spaying on growth rate. In his experiments there was no difference between the spayed and control females in growth rate. He used however only six control and six spayed animals, but noted their food consumption which also showed no difference between the groups. In another small experiment, Stahl found that castrated males grew faster than control females but had a poorer food utilization.

Hammond (1932) records the relative weights of rams, wethers and ewes.at four months of age as

124, 110 and 100, but points out that the ram-wether comparison suffers because the rams were not checked in growth by the operation. It seems unlikely however that the operation could account for all the observed difference.

Ivanov and Gerčikov (1938) found that the final weight reached by spayed gilts is considerably greater than that of the controls, the average excess being 12 to 15 per cent. The amount required for 1 kg. of gain was 27 per cent less in spayed gilts. But owing to environmental variations, such as incidence of disease, the results are not very uniform, especially in individual cases.

Habu and Ishihara (1935) used 10 bull calves; 4 of them were castrated at six months of age, 4 at one year old, and 2 were kept as controls. They were killed after a fattening period of 48 days. There was no significant difference in growth, height at withers or food utilization. Steers produced better beef, and those castrated at six months were superior in finish to those castrated at one year old. In the castrated animals the fat was more finely marbled and the subcutaneous fat was more smoothly laid on than in the bulls. Thymus weight was found to be greater and thyroid weight smaller in the steers. For a period of only 48 days of fattening, large differences can hardly be expected in these results.

Richter (1936) collected data on 120 cows and reported that there was no significant difference between spayed and normals in respect of total milk yield, duration of lactation or fat content. The qualityof the beef and the growth in body weight were slightly improved, but he concluded that spaying of healthy cows was not a suitable method for raising milk and beef production.

On the whole, the results of the above mentioned investigations on farm animals do not lead to any far-reaching conclusions. In connection with growth rate and efficiency of food utilization as affected by the removal of gonads, the evidence is often too scant to be conclusive. Furthermore, it is doubtful whether all the experiments were conducted in a satisfactory way. In such experiments, there is a minimum requirement (dependending on the extent of uncontrolled variation) in the numbers of test and control animals, which should be comparable in all other respects. The length of the experimental period should also be sufficient to give a satisfactory results. Winters and McMahon (1933) believe that a four-month test feeding-period is as short as can be permitted to obtain estimates of individual feeding efficiency in cattle. Further, in these experiments, it is not always clear whether environment has been controlled as far as is desirable. The absence of statistical estimates of uncontrolled variation is a difficulty in interpreting data in which the effects of treatment are not always consistent, and the numbers of animals are small.

OBJECTS OF THE EXPERIMENT.

The objects of the present experiment are as follows:

(a) To re-investigate the effects of gonadectomy in rats upon the efficiency of food utilization, and to observe the extent to which any difference between normal males and females in economy of food utilization is reduced by gonadectomy.

(b) To determine whether the age at which the operation is performed affects the final results of the test period of growth.

(c) To observe the growth of animals and of the various parts of the body to determine whether the operation affected the proportions which the parts bear to each other and to the whole. Four sets of comparisons were intended: (1) of normal males and females, (2) of castrated and control males, (3) of spayed and control females, and (4) of castrated males and spayed females. In comparing the growth of organs, special attention was to be paid to the endocrine glands, and to the urogenital organs.

(d) To discover the relationship existing between rate of growth and efficiency of food utilization.

EXPERIMENTAL MATERIAL AND TECHNIQUE.

Experimental animals.

The albino rats used in this experiment were selected from the stock animals of this laboratory, which were originally supplied by the Wistar Institute. The colony has arisen from two pairs of Wistar albinos and by the system of inbreeding adopted a relatively homogenous stock has been obtained.

In this experiment, seventy-five rats were used, comprising 34 males and 41 females. They were from ten litters distributed as shown in Table 1, which shows that approximately half the animals in each litter and of each sex were used as controls for the other half on which gonadectomies were performed.

The term gonadectomy refers to the removal of gonads in both sexes. Castration and spaying refer to the removal of testes in the males and ovaries in females respectively; in the latter case, the term ovariectomy is also used. Although both test and control animals have been operated on, in what follows the term "operated" refers only to gonadectomised rats.

The rats have been subdivided into two other major groups according to the age at which they were gonadectomised. Group A consists of those which were gonadectomised shortly after birth together with their controls. It includes six litters, Nos.6 and 8 operated on the day of birth, and Nos. 1, 5, 7 and 10 operated on at two weeks of age. Group B rats were gonadectomised at puberty, as fixed by the time of the opening of the vaginal orifice in the females. The average age at this time was observed to be 41 days and the operations were made on the 45th days of age. The litters concerned were Nos.2, 3, 4 and 9. The total number of animals in Group A was 42, and in Group B, 33.

Table 1.

Distribution of Animals Used in the Experiment.

Litter No. & Group	Male Rats		Female Rats		Total
	Control	Castrated	Control	Spayed	
1 A	2	2	2	2	8
2 B	2	2	2	2	8
3 B	2	2	1	1	6
4 B	3	3	3	2	11
5 A	1	2	2	2	7
6 A	2	1	1	2	6
7 A	2	2	3	1	8
8 A	1	0	2	4	7
9 B	2	1	3	2	8
10 A	1	1	2	2	6
Total	18	16	21	20	75
Group A:	9	8	12	13	42
Group B:	9	8	9	7	33

Feeding.

All the test as well as the control animals were fed the same food. The staple diet used was a synthetic one comprising : wheat offal, ground wheat, Sussex ground oats, ground barley, ground maize, meat and bone meal, dried milk, white fish meal, dried yeast, sodium chloride and cod-liver oil. The nutritive of this diet is as follows:

Digestible true protein	16.1%
Digestible protein equivalent	16.7%
Starch value	66.6%
Calcium	1.18%
Phosphorus	1.0%

This food has been extensively used at the Rowett Research Institute, Aberdeen, and this Institute, and has been found very satisfactory as far as growth, health and reproduction are concerned.

The method of feeding was a very troublesome matter before the start of this experiment. Various ways had been tried without much success. It was found that rats always like to hold the food in their paws when eating it. When cubes were fed in a wire hopper some of the rats carried away all they could and stored them at the corner of the cage.

T hey were soon mixed with the sawdust and soiled with water and urine. Paste was then tried and put into a glass jar. The only advantage of this method is that is prevents the food from being carried away, but there are more disadvantages. The rats used to sit in the jar, and soil the contents and further, the loss of water through evaporation made the percentage of dry matter contained in the paste vary all the time. The paste also provided an ideal environment for the growth of moulds.

Then it was decided that the food in the form of powder would be the best for obtaining the exact amount of food eaten by individual rats. The feeder used in this experiment was made with ordinary fruit tins cut to a height of 6.6 cm., with a diameter of 10 cm.. The food powder placed in the tin was covered with a lid slightly smaller than the tin in circumference, with a hole of 3 cm. in diameter in the center through which the rat could reach the food. Any food scrapped through the hole was retained on the lid and therefore easily returned.

The food in tins filled to 300 grams inclusive, was weighed and refilled twice a week. The difference in weight between the intervals of weighing represented the food consumed. Besides the stock diet, green vegetables, usually cabbage, cut into three square inches in size, were fed once a week.

The rats were found to be more active during the hours between 4 p.m. and 4 a.m. They ran about in the cages, and the bedding was frequently shifted about, consequently a certain amount of sawdust was thrown into the feeder. So it seemed advisable to take the feeder away in the evening and put it in next morning.

Water was provided in a porcelain dish, cleaned and refilled every morning.

Weighing.

The body weight of rats and food consumption were taken twice a week, but in this report only weekly weights are considered. Weights were recorded to the nearest gram.

Management.

Each rat was put into a separate galvanised wire cage, 16" × 18" × 10", with a label showing number, sex and date of feeding period. Straw or wood shavings and sawdust were used as bedding. The animals were kept in a well-ventilated hut, with automatically-controlled temperature.

Duration of the experiment.

The experimental feeding period of each litter lasted for ten weeks. For Group A animals this period began at 30 days of age, and for Group B at 48 days of age.

Technique of operation.

Before the operation the animals (both test and control rats) were earmarked. All the instruments were sterilised before use. Each animal was anaesthetised by inhalation of ether from cotton wool. The four limbs of the animal were tied up with bandage and pinned on an operating board. The hair near the site of incision on the animals body was clipped and antiseptics were applied.

The ovariectomy of female rats was as follows: Incisions were made on both flanks. The size of the incision dependended on the size of the animal, but was not more than 0.5 cm. in length. Ovaries and oviducts can readily be found with a pair of fine forceps just posterior to the kidneys. In older rats the ovaries were usually imbedded in fat, while in new-born ones a binocular microscope found helpful in locating the ovaries. The ovary and part of the Fallopian tube were severed with a pair of fine scissors. Bleeding was scarcely noticed. After the removal of the ovaries, the lumbar muscles were stitched or painted thinly with collodion, and the wound on the skin stitched with silk thread and a half curved needle. The control female rats were treated in the same way, except that the ovaries and Fallopian tubes were only examined and were left intact.

In male rats only one incision was made on the middle line of the abdomen in front of the sheath. Both testes and epididymides were brought forward through the inguinal canal and severed from their attachments. In older animals a ligature on the spermatic cord was always found necessary to prevent excessive bleeding from the spermatic vessels. Then the wound was stitched as in ovariectomy. The removal of epididymis does not make any difference to the effect of castration, Since Lawless (1936) demonstrated that the castration leaving the epididymes intact produced the same results as castration with removal of the epididymes.

Some difficulty was experienced in performing the operation on day-old rats owing to the small size of the organs and the frailness of the tissue. Worse still was the fact that some nursing mothers refused to suckle their offspring or even ate them, causing a certain mortality among the operated animals. In rats operated on or after two weeks of age death due to operation has not occurred.

Immediately after operation, any trace of blood on the young rat should be throughly wiped off with cotton wool. It is also advisable to keep the treated rats at a higher temperatur e (by wrapping them in cotton wool or exposing them to an electric radiator) for sometime before returning them to the nest. This seems to making it easier for them to revive their strength as well as to dry up the wound sooner, and render it less likely that the mother will kill them.

Technique of autopsy.

After the last body weight was taken, the animals were killed with ether. The nose-anus length and tail length were measured. Removing the head at the point just behind the cranium and the hyoid bone, its weight was recorded, and then that of the eyeballs. After the cranial roof had been removed, the brain was exposed and extracted by clipping the cranial nerves. The hypophysis could then be removed from the sella turcica. The weight of submaxillary and thyroid glands were next taken.

The petitoneal cavity was then opened, and the weights of the reproductive organs (testes, epididymides and other accessory genital organs in male rats; or ovaries and uterus in female rats) taken. The intestines, including contents, with mesentary and pancreas were removed and weighed together. Empty intestines alone were next weighed. Spleen and stomach, with and without contents, kidney and suprarenal glands were removed and weighed in order. In the thoracic cavity the thymus is found near the mediastinum. Heart and lungs were removed separately. The integument was next taken off and from it the subcutaneous as well as the gross visible fat in the abdomen were removed as completely as possible. The skeleton and musculature, including the spinal cord, were weighed together. The bones were macerated with 0.1 per cent KOH at 56° C. for 24 hours and then throughly washed with running water, kept dry for later measurement and examination.

In order to avoid drying, all dissected organs were preserved in covered glass dishes containing moist filter paper, and the weights were taken as soon as possible. The head, skeleton, musculature and spinal cord, stomach, intestines and fat were weighed on a platform balance, sensitive to the tenth of a gram. The other small organs were weighed in glass containers on an analytical balance, sensitive to the tenth of a milligram.

Statistical analysis of the data.

When working with biological material the difficulty is often met that a certain amount of variability even within groups of apparently similar animals will always be present. This renders it difficult to distinguish between differences due to a given treatment and those due to uncontrolled causes and raises the necessity of comparing groups of animals rather than individuals. Thus, when it is desired to compare the length or weight of certain parts or organs in two groups of animals, the mere comparison between the means of the two groups is not sufficient. It has to be tested whether a difference such as that observed might not arise through the mere chance of random sampling from a larger normally distributed population to which the two groups might belong.

Methods of making such statistical tests of the significance of the differences between means have long been known. The one used in this study has been described by Fisher (1936) and Snedecor (1938). An example from the data is given below to illustrate the way in which the calculations were made.

Table 2. (on Page 29)

Table 2 shows that the mean body length of control male rats was 22.26 cm., while that of castrated males was 21.69. Individual values, however, show overlapping of each group with the other, so that it is not possible to know from the means whether their difference is due to operation. A test of significance has therefore to be carried out, and has been given in detail in the next table.

The analysis at the bottom of the table shows that the ratio F of the mean square between groups to the mean square within groups (which serves as the estimate of the error) is 2.64. According to the tables of Snedecor (1938), the 5% point for degrees of freedom 1 and 32, is 4.15. Since 2.64 is smaller than this value, it must be concluded that the means of the two groups could have arisen by change more than once in 20 trials, and must therefore be considered insignificant on these data.

Table 2

Test of significance of difference in body length in cm. of castrated and control male rats

Observed lengths							
Control				Castrated			
22.4	21.6	23.6	22.4	22.4	21.8	21.5	22.7
22.3	22.5	24.3	21.4	20.7	21.0	21.0	24.5
23.0	23.6	22.1	22.0	20.0	21.4	21.5	21.0
23.0	21.5	20.7	21.4	22.5	20.8	22.2	22.1
22.2	20.7						

	Control	Castra ted	Total
Sum	400.7	347.1	747.8
Number of rats	18	16	34
Mean	22.2611	21.6938	21.9941
Sum of squared obs.	8936.43	7546.63	16483.06
C.F.,(Total mean)	8920.02	7529.92	16447.19
Sum of sq.deviations	16.41	16.71	35.87

Analysis of variance

Source of variance	D.F.	Sum of squares	Mean square	F obs.	F 5% point
Total	33	35.87	1.09		
Between groups	1	2.75	2.75	2.64	4.15
Within groups(error)	32	33.12	1.04		

The observed value of F is 2.75/1.04 according to Snedecor (1938).

EXPERIMENTAL DATA.

1. Growth rate.

During the experimental feeding period of ten weeks following the gonadectomy of the test animals, weighings of all the rats were made twice weekly. In Table 3, the differences between the mean initial and final weights for each group have been brought together.

Table 3.

Gain in Weight in Grams during the Experimental Feeding Period of 10 Weeks.
Number of rats in brackets.

	Males			Females	
	Group A	Group B		Group A	Group B
Control	169.1 (9)	134.8 (9)	Control	113.2 (12)	87.3 (9)
Castrated	151.0 (8)	123.5 (8)	Spayed	130.0 (13)	109.0 (7)
Difference	-18.1	-11.3		+16.8	+21.7 ( )

It is evident from this table that the castration of male rats has reduced the gain in weight as compared with the control males irrespective of the time of castration. Among the females the effect of gonadectomy has been different. The spayed animals are heavier in both groups than their controls. Statistical treatment of these data revealed that the effects of gonadectomy were significant. In the males, the difference was significant at the 5% point, and in the females at the 1% point.

The influence of age at the time of castration or spaying appears from Table 3 to require further investigation. In the males the difference between the controls and the castrated in Group A is larger than the corresponding difference in Group B in which the castration took place later. If it is supposed that the absence of testes is detrimental to growth, then the longer the absence the greater the difference. The observed results would agree with this supposition. But in the females the difference is greater in the Group B animals, and is thus contrary to the expected results. The numbers of animals available for this comparison now appears however too small to be decisive for this question.

Comparing now the males with the females, it will be seen that both castrated and control males exceeded in growth rate the spayed and control females. It is true that the initial weight of the females was smaller than that of the males, yet the gap between the two growth curves became greater with advancing age. The difference between the control males and females has been observed by all workers to favour the males. It is of more interest to compare the gonadectomised animals. The removal of the gonads should tend to eliminate sex differences insofar as these depend on the functioning of the gonads. The removal of the testes retards weight increase while removal of the ovaries accelerates it so that gonadectomy causes the growth curves to become more similar than those for normal males and females. Reference to Figures 1 and 2 will show how the sex difference, although very much reduced by gonadectomy, is not completely removed. The remaining difference is still in favour of the males and is expressed principally during the first two to four weeks after the beginning of the test period (Fig. 3 and 4). Thereafter the difference in both groups appears to have been very slight.

As judged by the mean increases in weight, the early castration and spaying has been no more effective in reducing the sex difference than the later. The difference between the control males and females has been reduced by about 5/8 by the early gonadectomy, and by a little under 6/8 by the later.

So far as the present data permit drawing conclusions it seems that the sex difference in rate

Figure 1.

Growth Curves of Control Rats

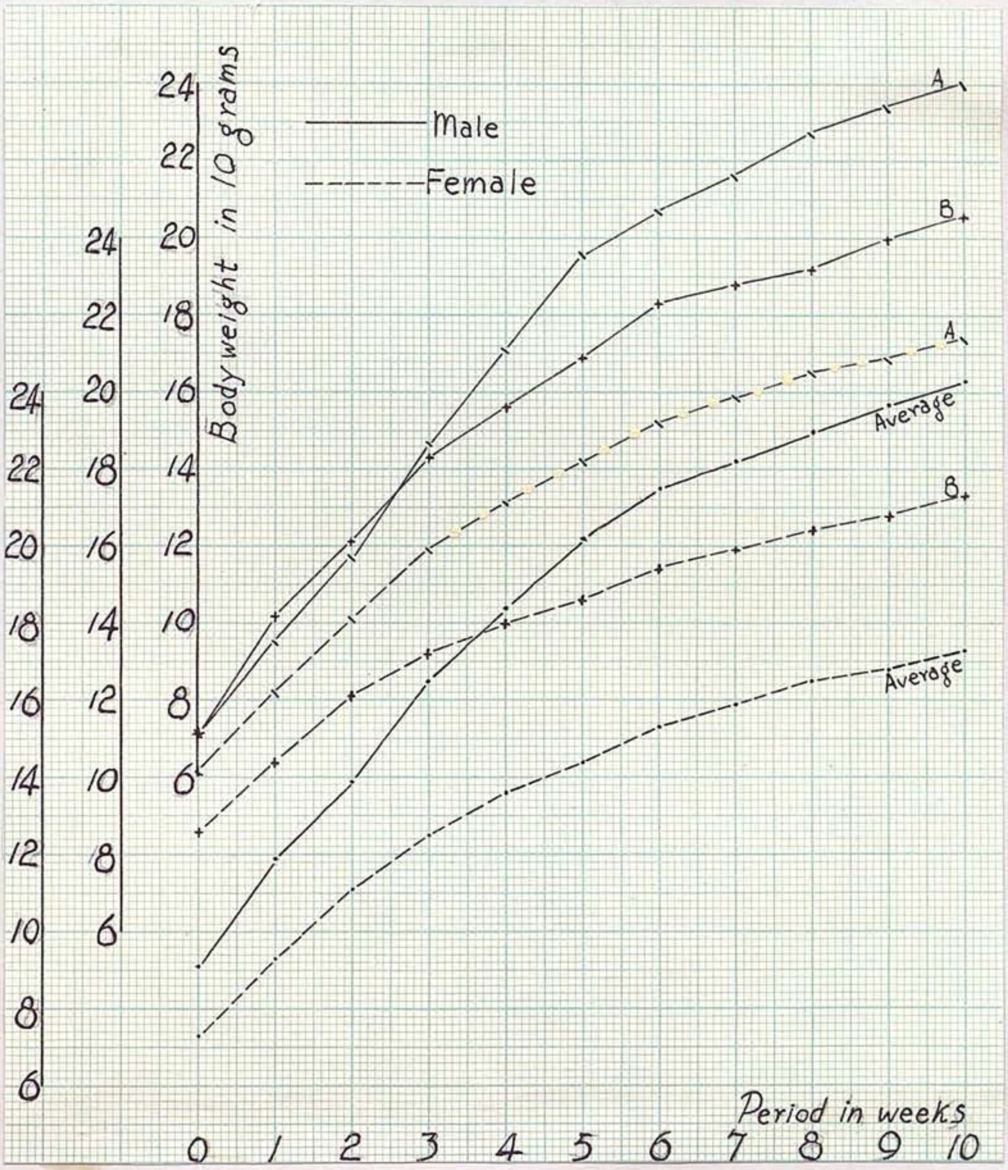


Figure 2.

Growth Curves of Gonadectomised Rats.

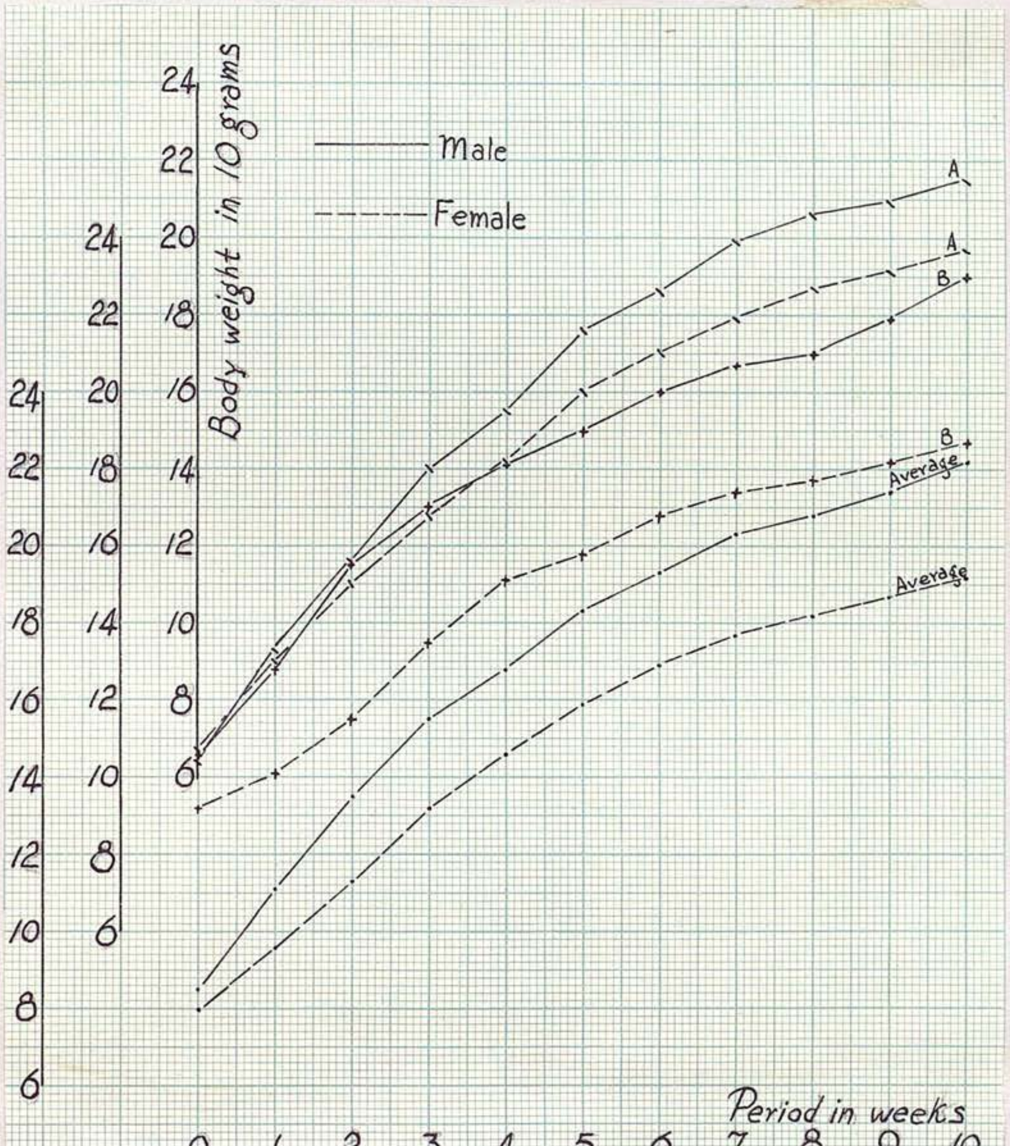


Figure 3.

Growth Curves of Male Rats.

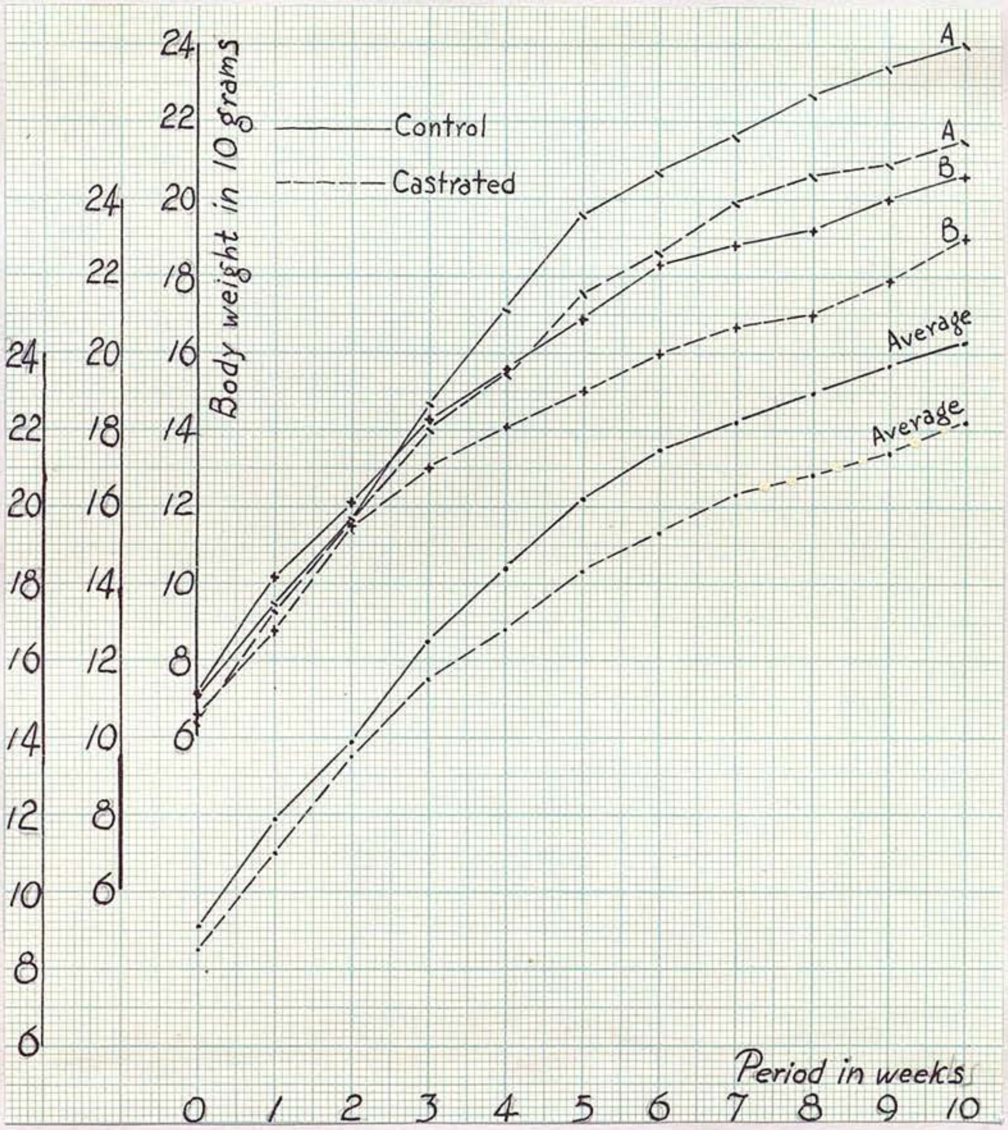
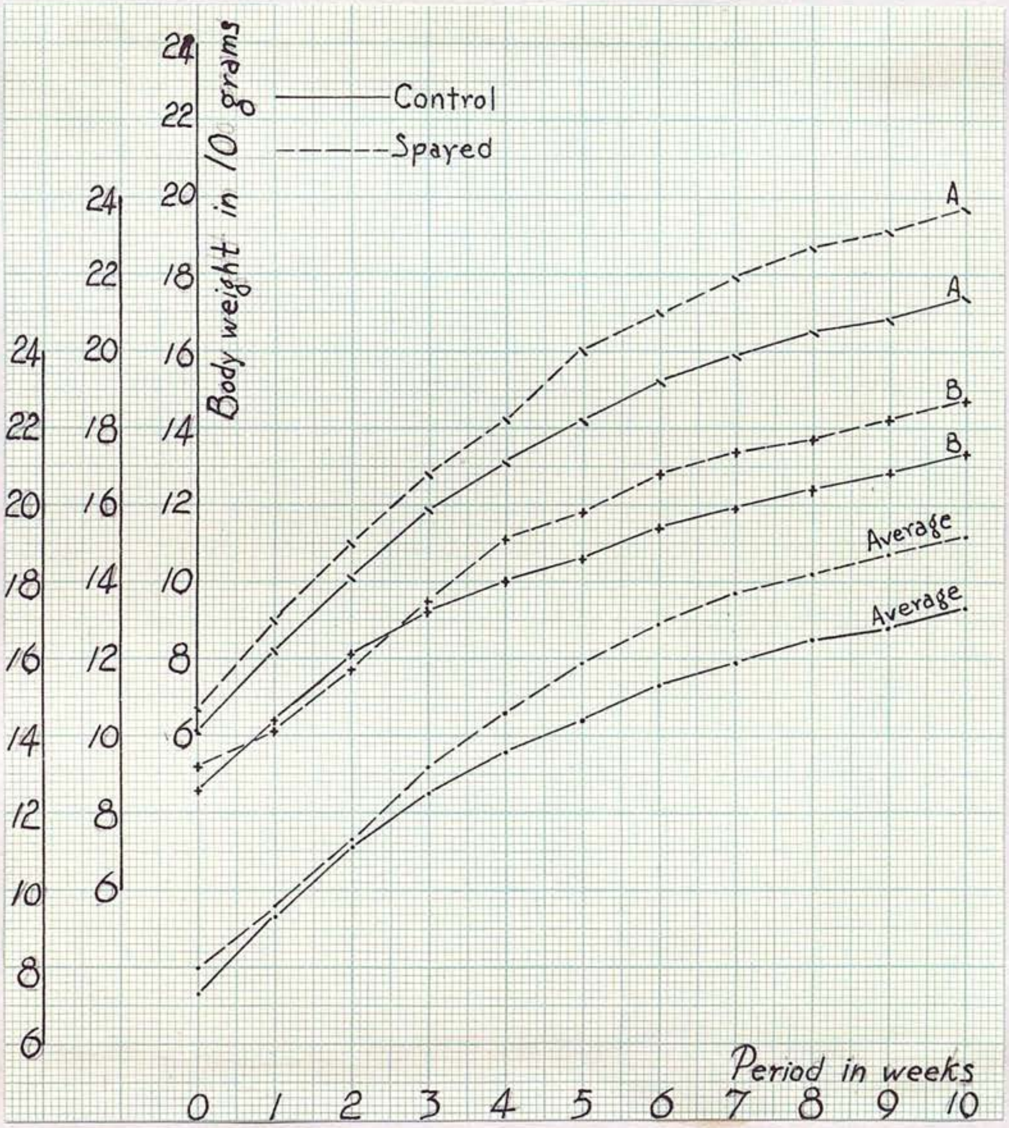


Figure 4.

Growth Curves of Female Rats.



of growth cannot be entirely eliminated by gonadectomy even as early as one day old. Since the experiment did not proceed beyond the ten weeks feeding period, nothing can be said of the sex differences beyond this time.

2. Food consumption.

The amount of food consumed during the ten weeks feeding period was recorded and in Table 4 are given the mean quantities consumed per rat for each of the eight sub-groups of animals.

Table 4.

Food Consumption in Grams during the 10 Weeks Feeding Period. Number of animals in brackets.

	Males			Females	
	Group A	Group B		Group A	Group B
Control	1189 (9)	1160 (9)	Control	1042 (12)	1038 (9)
Castrated	1089 (8)	1068 (8)	Spayed	1031 (13)	971 (7)
Difference	-100	- 92		- 11	- 67

It will be observed that in all cases the control rats ate more than the gonadectomised. Reference to Tables 3 and 4 will show that the control males exceeded the castrated males in weight and in food consumption in approximately the same degree. Thus the effect of castration of efficiency as judged by mean gain in weight in relation to food consumption appears to be small in these rats. The food consumption of the control females, however, is higher than that of the spayed, in spite of the fact that they put on less weight. Here one may suspect that spaying has improved the efficiency of food utilization.

A comparison of the amount of food eaten by males and females may be made with the aid of Figures 5, 6, 7 and 8. Here are shown the cumulative food consumption during the 10-week feeding period. Figure 5 shows that the control male rats have eaten about 130 grams more than the control females, and this amount has been accumulated during the whole period. Figure 6 compares the gonadectomised males and females. Here the males have also eaten rather more than the females throughout, but the difference is much less than that found between the control males and females.

Figure 5.

Cumulative Food Consumption of Control Rats.

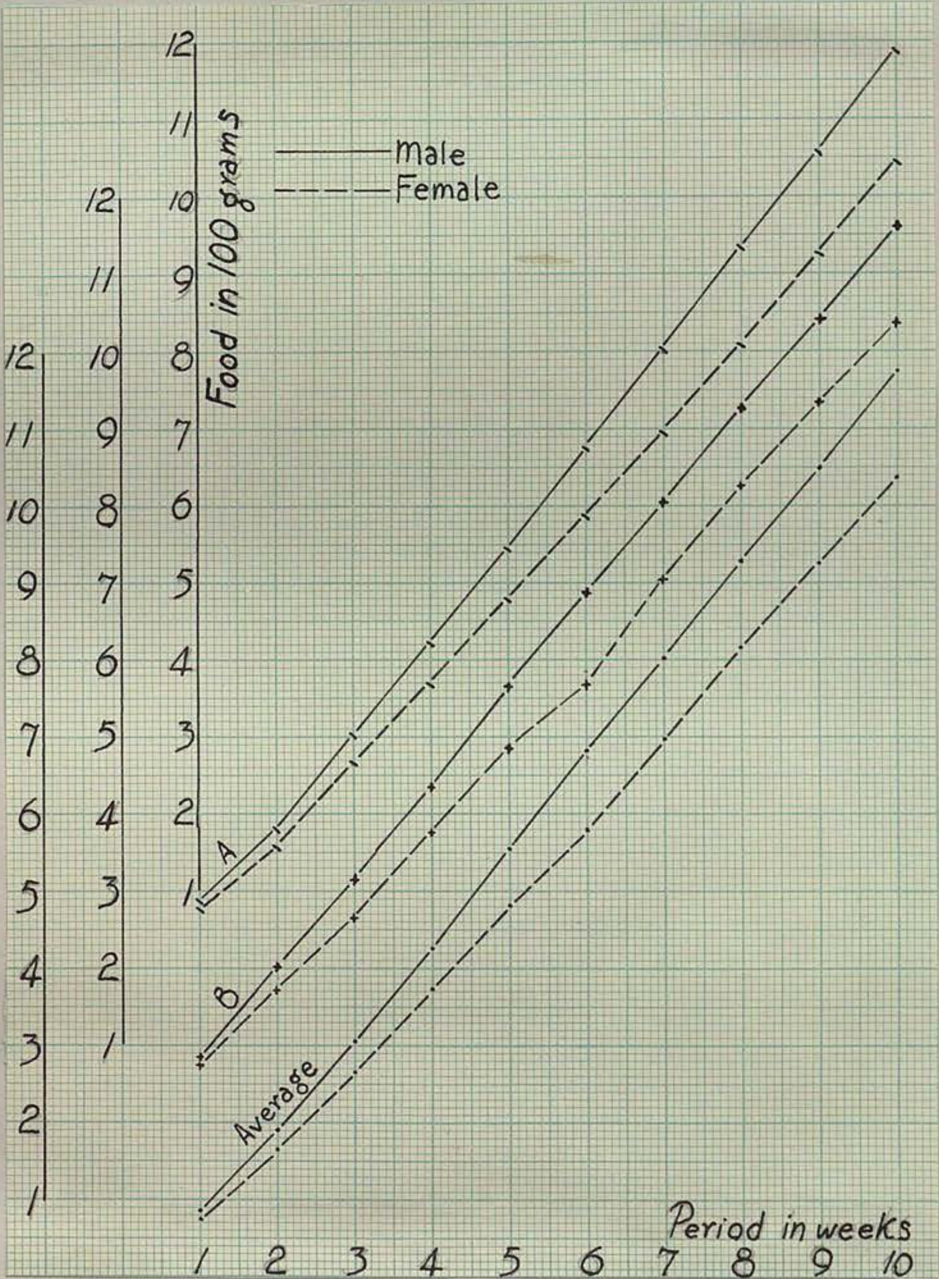


Figure 6.

Cumulative Food Consumption of Gonadectomised Rats.

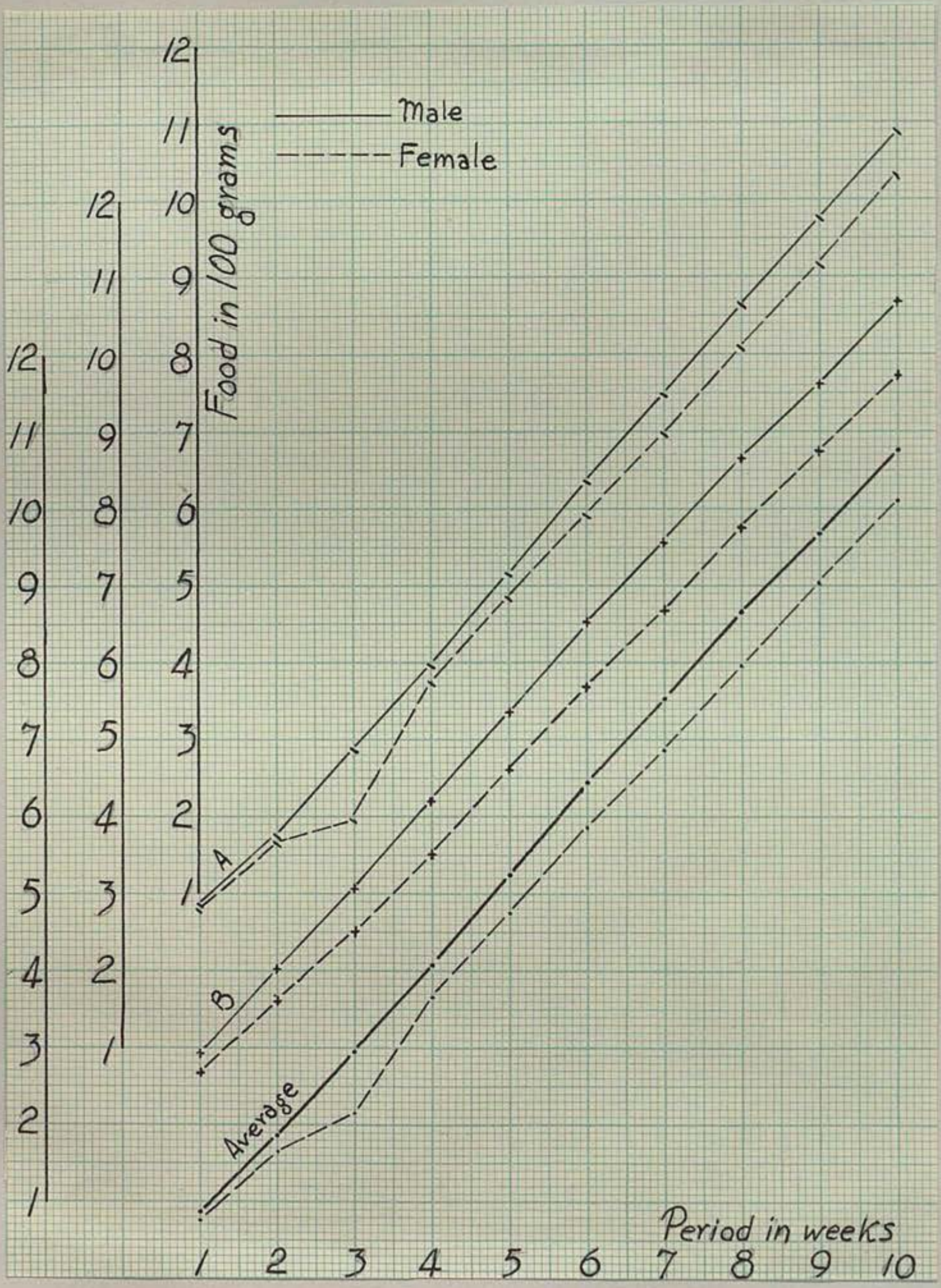


Figure 7.

Cumulative Food Consumption of Male Rats.

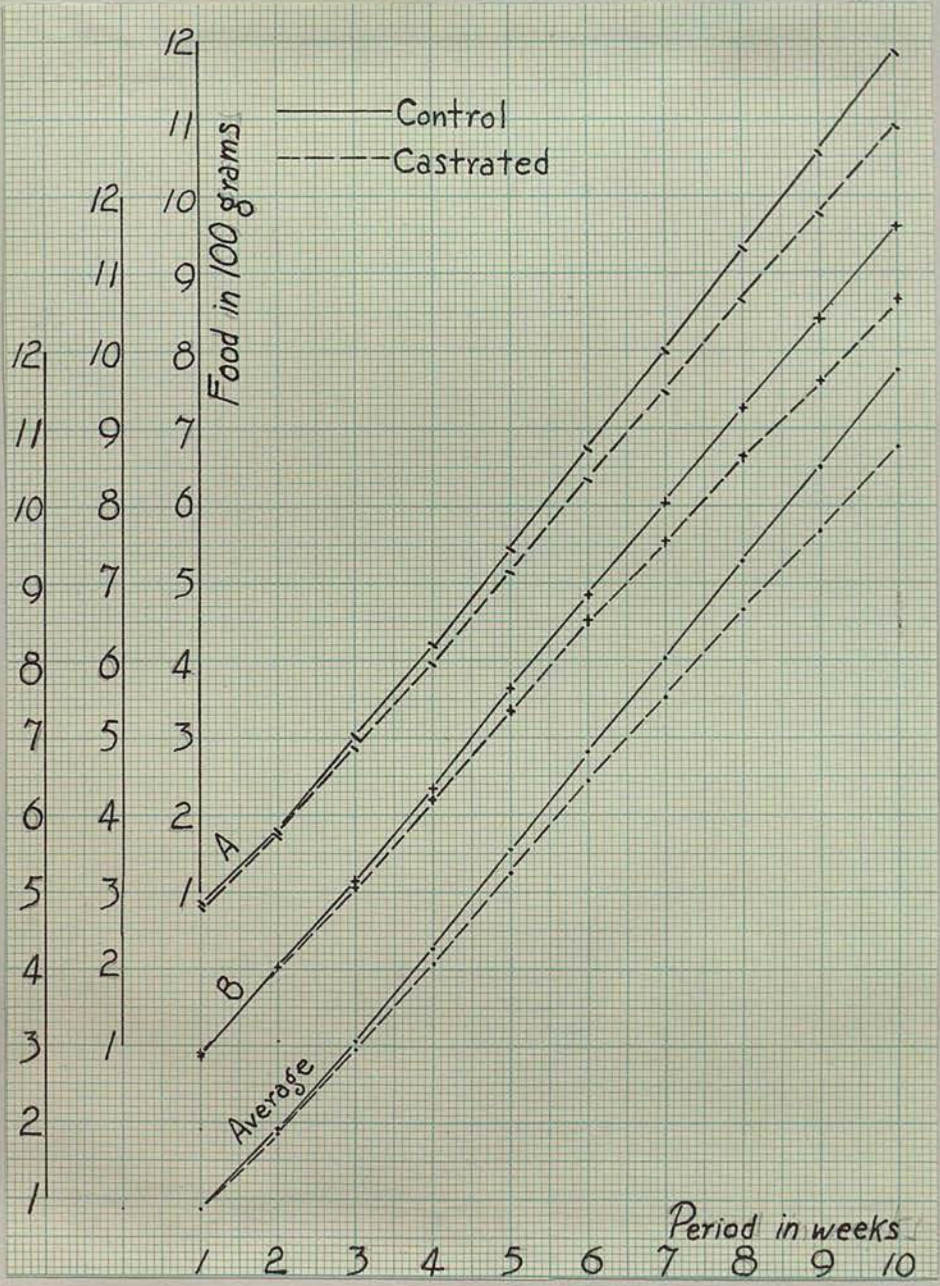
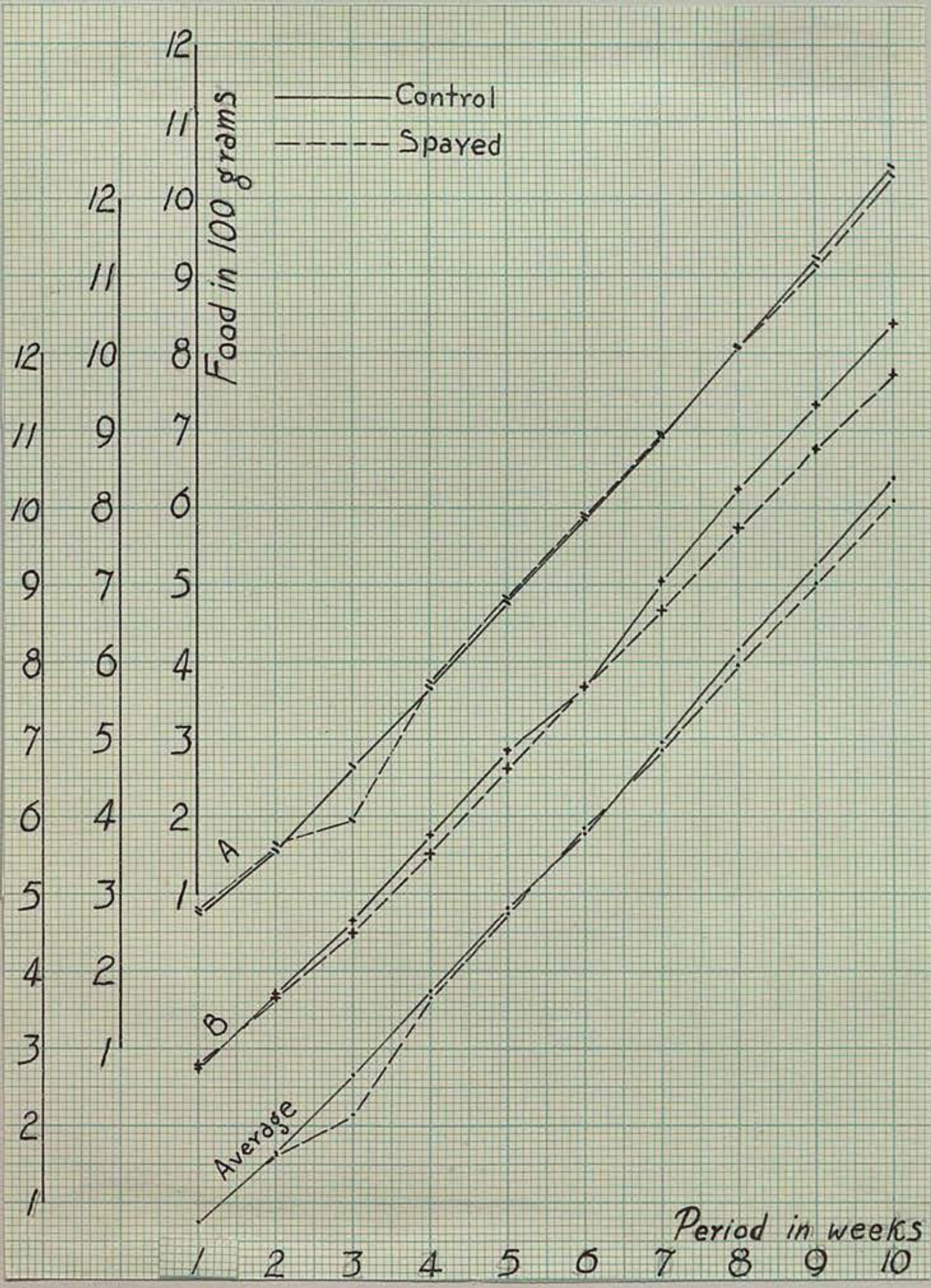


Figure 8.

Cumulative Food Consumption of Female Rats.



3. Efficiency quotient.

The calculation of the efficiency quotient was made according to the formula of Winters and McMahon (1933):

$$E.Q. = \frac{\text{Gain in weight(gr.)}}{\text{Food consumed(gr.)}} \cdot \frac{(\text{initial weight}+\frac{1}{2}\text{ gain})}{100}$$

The object of the calculations was to arrive at an estimate of efficiency of food utilization which would allow for the varying total live weight.

The results of these calculations are summarised in Tables 5 and 6. The figures given in them show the mean weekly values of the E.Q. for each sub-group of rats. Consideration of them shows that the differences week by week between the castrated and control males are not consistent in sign nor magnitude and it must be assumed that the difference in efficiency as measured in this way cannot be large. This conclusion was foreshadowed by the information given in Tables 3 and 4. In Table 6, however, the difference in E.Q. between spayed and control females is in the same direction in the bulk of the paired values. Some confidence may be felt therefore that the spaying has resulted in a greater efficiency in the utilization of food.

Table 5

Mean Efficiency Quotients for male rats for each week of experimental feeding

Week	Group A		Group B		Groups A and B	
	Contr.	Castr.	Contr.	Castr.	Contr.	Castr.
1	0.228	0.287	0.460	0.276	0.344	0.282
2	0.259	0.252	0.248	0.358	0.254	0.305
3	0.328	0.269	0.341	0.241	0.335	0.255
4	0.311	0.203	0.201	0.165	0.256	0.184
5	0.367	0.307	0.196	0.144	0.281	0.225
6	0.170	0.188	0.255	0.190	0.213	0.179
7	0.163	0.183	0.101	0.130	0.132	0.157
8	0.172	0.112	0.072	0.060	0.122	0.086
9	0.136	0.057	0.150	0.185	0.143	0.121
10	0.106	0.107	0.129	0.230	0.118	0.169
Mean	0.224	0.196	0.216	0.198	0.220	0.196

Table 6

Mean Efficiency Quotients for female rats for each week of experimental feeding

Week	Group A		Group B		Groups A and B	
	Contr.	Spayed	Contr.	Spayed	Contr.	Spayed
1	0.207	0.227	0.233	0.135	0.220	0.181
2	0.208	0.233	0.194	0.185	0.211	0.209
3	0.181	0.198	0.147	0.259	0.166	0.228
4	0.148	0.182	0.101	0.215	0.124	0.199
5	0.131	0.258	0.086	0.102	0.109	0.180
6	0.139	0.157	0.140	0.153	0.139	0.155
7	0.103	0.150	0.053	0.096	0.078	0.123
8	0.082	0.121	0.072	0.061	0.077	0.091
9	0.051	0.081	0.061	0.077	0.056	0.079
10	0.078	0.098	0.075	0.102	0.077	0.100
Mean	0.133	0.170	0.116	0.139	0.125	0.155

To permit easy appreciation of the relative sizes of the difference in efficiency, Table 7 has been compiled. It shows that average weekly superiority of the male controls over the female controls was 43 per cent. Castration and spaying reduced this to 19 per cent. Similarly, control males were 7 per cent more efficient than the castrated, and the control females 31 per cent less efficient than the spayed. The regularity with which the males have a higher E.Q. than the corresponding females leaves no doubt that they are more efficient either as normal or as castrated males.

There is a tendency apparent in all eight sub-groups for the rats to become less efficient as they grow older and their rate of growth diminishes. This is to be expected since their food consumption remains comparatively constant while their maintenance requirements increase.

Table 7

Percentage differences in Efficiency Quotient for each week of experimental feeding

Week	Males	Females	Control	Gonadectomized
	C - G	C - G	♂ - ♀	♂ - ♀
1	18	18	36	36
2	-20	- 4	21	31
3	24	-39	51	11
4	28	-60	52	- 8
5	20	-66	61	20
6	16	-11	35	13
7	-18	-58	41	21
8	30	-12	37	- 5
9	16	-43	61	34
10	-43	-30	35	41
Mean	7	-31	43	19

C, control; G, gonadectomized.

4. Growth of the body and organs.

After the 10-weeek feeding period was finished, all the rats were autopsied in the way described on p.25-27. The data obtained have been studied both as absolute and relative measurements. The object has been to determine whether the effects of gonadectomy on the growth of the rats have been produced by a general hastening or retarding of body growth, or whether particular organs have been affected more than others.

Since the final weights of the animals in Groups A and B were very similar (see Fig. 1-4, p.33-36), the groups have been combined in the following sections of the work. The effect of including the slight group differences is to increase the variability a little within sexes and treatments. The mean actual and relative weights and lengths are recorded in the following tables. Table 8 gives the means, coefficients of variation, and the significance of differences between control and castrated rats. Table 9 gives the same data for females. These tables may be considered along with Table 10, which gives the differences between males and females, and their significance.

Table 8

Comparison of carcass measurements of castrated and control male rats. Weights in grams and lengths in centimetres.

Organ or part	18 Control rats Mean Actual	Relative	C.V.	16 Castrated rats Mean Actual	Relative	C.V.	Difference Actual	% of control	F	Significance
Total body weight	235.2	100	7.4	214.6	100	9.6	-20.6	-8.8	10.0	SS
Nose-anus length	22.26	9.46	4.4	21.69	10.11	4.9	- 0.57	-2.6	2.6	NS
Tail length	18.18	7.73	7.3	17.91	8.35	7.0	- 0.27	-1.5	<1	NS
Head weight	19.19	8.16	9.2	18.09	8.43	7.3	- 1.10	-5.7	<1	NS
Skeleton, muscle and spinal cord	132.2	56.19	9.3	124.4	57.97	11.2	-7.8	-5.9	3.0	NS
Integument	31.74	13.49	12.3	29.93	13.95	15.7	- 1.81	-5.6	1.5	NS
Fat	8.54	3.63	45.5	8.91	4.15	48.6	+ 0.37	+4.2	<1	NS
Brain weight	1.688	0.717	7.9	1.659	0.773	6.8	- 0.029	-1.7	<1	NS
Hypophysis weight	0.011	0.0047	21.1	0.014	0.0066	15.9	+ 0.003	+26.6	13.8	SS
Eyeballs	0.207	0.088	9.7	0.202	0.094	13.9	- 0.005	-2.5	<1	NS
Submaxillary gland	0.503	0.214	11.2	0.473	0.220	14.7	- 0.030	-5.9	1.9	NS
Thyroid gland	0.018	0.0075	18.7	0.018	0.0083	23.9	+ 0.000	+1.8	<1	NS
Thymus gland	0.218	0.093	26.3	0.318	0.148	15.2	+ 0.101	+46.3	30.2	SS
Heart weight	1.199	0.510	14.4	1.145	0.534	16.7	-0 .054	-4.5	<1	NS
Lungs weight	2.146	0.91	28.7	2.255	1.051	24.6	+ 0.109	+5.1	<1	NS
Liver weight	12.39	5.27	9.6	10.36	4.83	11.8	- 2.03	-16.6	24.1	SS
Spleen weight	0.839	0.356	24.8	0.882	0.411	27.5	+ 0.043	+5.1	<1	NS
Suprarenal weight	0.042	0.018	12.6	0.049	0.023	21.3	+0.007	+16.6	6.3	S
Kidneys weight	1.117	0.475	10.9	1.769	0.824	11.6	+0.652	+18.6	27.6	SS
Stomach & contents	3.289	1.398	43.1	2.567	1.196	44.8	-0.722	-22.0	2.6	NS
Stomach, empty	1.117	0.480	11.5	1.190	0.550	18.2	+o.073	+6.5	1.5	NS
Intestines with contents	18.02	7.66	14.6	17.01	7.93	13.9	-1.01	-5.6	1.4	NS
Intestines, empty	5.75	2.44	27.3	5.45	2.54	19.1	-0.30	-5.3	2.2	NS
Testes weight	2.073	0.881	9.5							
Epididymes weight	0.871	0.370	30.2							
Other ♂ accessories	1.564	0.665	36.7	0.069	0.032	63.6	-1.495	-95.6	101	SS

C.V., coefficient of variability in %. The 5% and 1% points of F are 4.15 and 7.5 respectively.

Table 9

Comparison of carcass measurements of spayed and control females. Weights in grams and lengths in centimetres.

Organ or part	21 Control rats Mean Actual	Relative	C.V.	20 Spayed rats Mean Actual	Relative	C.V.	Difference Actual	as % of control	F	Significance
Total body weight	165.6	100	8.9	185.5	100	7.9	+19.9	+12.0	18.6	SS
Nose-anus length	20.24	12.23	4.3	20.71	11.16	3.3	+ 0.47	+ 2.3	3.5	NS
Tail length	16.54	9.99	7.8	17.70	9.54	7.1	+ 1.16	+ 7.0	8.5	SS
Head weight	14.86	8.98	6.3	16.48	8.88	8.6	+ 1.62	+10.9	18.6	SS
Skeleton, muscle and spinal cord	90.00	54.36	10.7	104.20	56.16	9.1	+14.2	+15.8	22.7	SS
Integument	21.37	12.91	14.1	25.12	13.54	14.1	+ 3.75	+17.6	13.3	SS
Fat	8.16	4.93	57.4	10.03	5.41	38.1	+ 1.87	+22.5	1.9	NS
Brain weight	1.566	0.946	6.2	1.618	0.870	7.5	+ 0.052	+ 3.3	2.3	NS
Hypophysis weight	0.0118	0.007	19.0	0.0126	0.007	17.8	+ 0.0008	+ 7.1	1.5	NS
Eyeballs weight	0.163	0.098	18.1	0.174	0.094	16.8	+ 0.011	+ 7.2	1.6	NS
Submaxillary gland	0.390	0.236	15.3	0.416	0.224	14.1	+ 0.026	+ 6.6	1.9	NS
Thyroid gland	0.0161	0.010	14.1	0.0158	0.009	12.7	- 0.0003	- 1.9	<1	NS
Thymus gland	0.2262	0.137	25.1	0.2969	0.160	22.5	+ 0.0707	+31.3	13.3	SS
Heart weight	0.815	0.492	15.5	0.948	0.511	13.8	+ 0.133	+11.4	5.6	S
Lungs weight	1.785	1.080	20.6	1.954	1.052	17.4	+ 0.169	+ 9.5	2.3	NS
Liver weight	8.81	5.32	10.9	9.22	4.97	11.3	+ 0.41	+ 4.6	1.6	NS
Spleen weight	0.711	0.429	28.7	0.721	0.389	17.3	+ 0.010	+ 1.4	<1	NS
Suprarenal glands	0.066	0.040	16.1	0.059	0.032	18.5	- 0.007	-11.5	4.9	S
Kidneys weight	1.615	0.975	9.7	1.665	0.897	8.7	+ 0.050	+ 3.1	1.1	NS
Stomach & contents	2.603	1.572	42.2	2.871	1.547	43.0	+ 0.268	+10.3	1.8	NS
Stomach, empty	0.928	0.561	11.7	1.071	0.577	20.6	+ 0.143	+15.4	7.0	S
Intestines & contents	16.27	9.83	10.7	17.14	9.24	11.8	+ 0.87	+ 5.4	2.2	NS
Intestines, empty	4.52	2.73	24.9	4.80	2.59	26.0	+ 0.28	+ 6.2	1.8	NS
Ovaries	0.074	0.045	28.1							
Uterus	0.469	0.283	37.1	0.066	0.036	67.7	- 0.403	-85.9	101	SS

C.V., coefficient of variability in %. The 5% and 1% points of F are 4.09 and 7.33 respectively.

Table 10

The significance of differences between means for males and females expressed as percentages of means for males

Organ or part	Controls		Gonadectomized	
	% Diff. ♂♂ - ♀♀	Signif.	% Diff. ♂♂ - ♀♀	Signif.
Total body weight	29.61	SS	13.60	SS
Nose-anus length	9.07	SS	4.52	SS
Tail length	9.02	SS	1.17	SS
Head weight	22.56	SS	8.90	SS
Skeleton, muscle and spinal cord	46.94	SS	16.22	SS
Integument	32.67	SS	16.07	SS
Fat	4.19	NS	-12.62	NS
Brain weight	7.23	SS	2.47	NS
Hypophysis weight	- 5.85	NS	10.38	NS
Eyeballs	21.56	SS	13.76	SS
Submaxillary gland	22.42	SS	12.05	S
Thyroid gland	9.60	NS	11.91	NS
Thymus gland	6.75	NS	- 3.90	NS
Heart weight	28.97	SS	17.14	SS
Lungs weight	13.35	NS	16.82	S
Liver weight	11.10	SS	28.88	SS
Spleen weight	15.19	NS	18.21	S
Suprarenal glands	-19.45	S	-57.37	SS
Kidneys	25.68	SS	5.88	NS
Stomach & contents	-11.84	NS	20.86	NS
Stomach, empty	16.92	SS	-10.00	NS
Intestines & contents	9.71	S	- 0.76	NS
Intestines, empty	21.41	SS	11.92	NS

SS, significant at the 1% point; S, significant at the 5% point; NS, non-significant.
Weights in grams and length in centimetres.

Body weight:- Castrated male rats in both groups were found to be lighter than their controls, the difference being significant at the 1% point. The same degree of certainty attaches to the differences found between the test and control females, between the male and female controls and the male and female test rats. These results are in agreement with the findings of Evans and Simpson (1927), van Wagenen (1928), Korenchevsky (1934) and of Lawless (1936). But Slonaker (1930) observed greater body weight in castrated male rats, while Stotsenberg (1909), Hatai (1913, 1915) working with albino rats and Masui and Tamura (1926) in mice reported that castration does not materially make any difference so far as the body weight is concerned. Holt, Keeton and Vennesland (1936) found overlapping results in their experiment and considered it inconclusive.

Nose-anus length (body length):- As pointed out by van Wagenen (1928), body length shows less response to castration than tail length, and it is therefore desirable to record nose-anus length and tail length separately.

The difference in actual nose-anus length between control males and females is highly significant with an F value of 46.3 which is six times greater than the expected 1% point. Gonadectomised

animals

animals have a difference of 4.5% favouring the males, about half of that between control males and females, but it is still significant. However, when the difference of body weight is taken into consideration, it shows that the control females, which are lightest in body weight as a group, have the greatest body length; on the other hand, the control males have the smallest relative length although heaviest of all. The order of absolute body length from greater to smaller is as follows: control males, castrated males, spayed females and control females, while the order for relative body length is just the reverse.

The castrated males failed to attain the body weight of the control males by 8.75%, yet they only failed to grow 2.56% in body length, which seems to suggest that the general skeletal growth of castrated males is not as much hindered by the operation as the body weight.

The spayed gained 12.01% more in body weight than their control sisters, yet they only surpassed them by 2.32% in body length. An illustration of the size difference between spayed and normal female rats is given in Fig. 9. Hatai (1915)

Fig. 9. Photograph illustrating the difference in body size of spayed (left) and control (right) female albino rats.

obtained very similar results. Masui and Tamura (1926) reported that normal female mice are longer in body length than males, and that the latter increase their length after castration, but spaying produces no effect on females. Stotsenburg (1913) noted longer body length in spayed rats. Moore found that the body length of normal female guinea pigs (1922) is greater than that of castrates and that normal females exceeded spayed ones. Greater body length was observed in spayed femal es rats by Freudenberger and Howard (1937) and Freudenberger and Hashimoto (1937), yet Freudenberger and Billeter (1935) could not find any significant change. In male rats van Wagenen (1928) stated that castrates attained a smaller body length than controls.

Relative weights and lengths of males and females are given in Tab. 8 and 9,p. 49 & 50.

Tail length:- It is interesting to note that males have longer tails than females, but relatively the females have the longer tails. A significant difference will be seen between control males and femal es, the latter being 9.02% smaller. After gonadectomy the difference is reduced to only 1.17%.

(to be continued on page 58)

(continued from page 55)

Castrated males are 1.49% smaller in tail length than controls, but this is not significant. Spayed females are significantly longer than controls, with a difference of 7.01%. Hatai (1913) found that castrated males were about 5% longer in tail length, but in females there was only a difference of 0.61% in favour of the spayed as compared with controls. In a later experiment Hatai (1915) reported a greater tail length (4.7%) in castrated rats than in controls, and in spayed females a slightly shorter tail than the controls. Freudenberger and associates (1935, 1937, 1937) reported greater tail length in controls.

Comparison of organ size in gonadectomised and control rats:- A general inspection of Table 8 shows that the castrated males were smaller than the control males in most of the large organs. The total body weight was significantly less, but the differences between the means for the various parts of the body with certain notable exceptions, were not significant. The general tendency for all the larger organs to share the reduced size in castrated animals is however clear. This tendency reaches a high level of significance in respect of the liver.

The most noteworthy feature of the table is the increased (and not lowered) size of the small organs of internal secretion, namely, the hypophysis, the thymus and the suprarenals. The average weights of these organs in the castrates were not only absolutely but relatively considerably larger than those of the controls. Further consideration to these facts will be given when the organs are dealt with individually.

With regard to the females in Table 9, the fact that the spayed rats were much larger than their controls has resulted in a greater number of significant differences being found in respect of the various organs. The larger size of the spayed rats is reflected in the larger size of most of the organs, but here again some notable departures from this rule occur which involve the endocrine glands. The thymus is again much enlarged, but the suprarenals are relatively much smaller than in the controls, and the weight of the hypophysis, although actually larger, is relatively the same weight in spayed and control females.

In contradistinction to the males, the liver shows little difference in actual weight between test and control females. This is also true for the kidneys which were relatively greatly enlarged in the castrated males.

Continuing the general review to Table 10, it will be seen that the male controls exceed the female controls in total body weight and also in most of the separate organs. The only exceptions are the weights of hypophysis and suprarenals which are actually smaller in the males, and relatively much smaller. It is also noteworthy that although actually larger in the males, the body and tail length, and the weights of brain, thyroid, thymus and fat are relatively much smaller (Tables.8 & 9). The extra weight of the males is clearly derived from greater development of muscle, head and integument.

Table 10 also gives a comparison of the gonadectomised rats. Associated with a smaller difference in total body weight there were smaller differences in the sizes of organs but on the whole the the comparisons run parallel to those between the control rats. The suprarenals again show distinctive behaviour being relatively much larger in the spayed females than in the castrated males.

The relative sizes of organs given in Tables 8 and 9 serve to bring out clearly the fact that the gonadectomised rats are very similar in the proportions of the chief structural features of the body to control rats of the same sex. It may however be noted that the castrated males, although lighter than the controls, are relatively longer; and that the spayed females although heavier, are relatively shorter than their controls.

The coefficients of variability shown in Tab. 8 and 9, confirm in general ~~with~~ previously published data. Body weight and length have fairly low variability. Most of the larger organs are of medium variability, while the lungs, spleen, fat, accessory male organs, female genitalia, thymus, and suprarenals are of high variability. This division is similar to that found by Eaton (1938) in guinea pigs. Freudenberger (1933), Lawless (1936) and Freudenberger and Hashimoto (1937) agree that the thyroid, thymus, lungs, spleen and suprarenals are very variable organs. There is also general agreement that variation in body length is very low.

The difficulty of separating the fat completely from its attachments reduces the value of the observed weights. Nevertheless, it is of interest to note that the high variability in amount of fat is consistent with what is experienced in larger animals of economic importance, and may also arise from genetic differences.

Head weight:- The difference of head weight between control males and females is 22.56% in favour of the males. The same relation holds after gonadectomy, but the difference is only 8.9%. Control males are not significantly heavier in head weight than castrates. In females, however, the difference between spayed and controls is even more significant than that between castrated males and spayed females. The order of relative weight is the reverse of that of absolute head weight. Freudenberger and associates also found that the head weight of the spayed rats was significantly greater than controls in three experiments.

Skeleton, muscles and spinal cord:- A striking difference in the weight of skeleton, muscles and spinal cord is exhibited between control males and females, the former being 46.94% heavier. This difference was reduced to one third among gonadectomised animals. Castrated males are 5.89% lighter than controls, but this is not considered significant. In females, spayed rats are significantly heavier, with a difference of 15.84% and F value of 22.72. In relative weight the males were practically the same as females. Castrated males and spayed females have greater relative weight than their respective controls, but the difference is slight. There seems no previous report on this weight.

Integument:- Control males have a greater integument weight (32.67%) than that for control females. Undoubtedly this difference is significant. The females are 16.07% lighter in their weight of integument than test males. Comparing weights within sex, control males are 5.61% heavier than castrates, while spayed females exceed by 17.55% the controls. The latter should be considered very significant, and confirms fully to the findings of Freudenberger and his colleagues.

The relative weights of integument of all four groups are very similar. Operated males and females are heavier in integument regardless of their body weight. The gross visible fat was removed before the weight of integument was taken.

Brain:- Brain weight of male controls is significantly greater than that of female controls. The percentage difference is 7.23. The difference between test males and females is insignificant, i.e. 2.47%. Castrates are 1.72% lighter, and spayed females 3.32% heavier than their respective controls.

Both are insignificant. Since brain is one of the chief components of head weight, naturally it follows the same order as the latter.

Hypophysis:- Among controls, the females have a 5.85% greater weight of hypophysis than males. Although it is not significant if only the absolute values are compared, however, since the females are much lighter in their body weight than males, the relative weight of hypophyses for females is much greater than that of males. Castrated rats were 10.38% greater in their hypophysis weight than spayed females, yet owing to wide variation this cannot be considered significant. Test males are 26.64% heavier than controls in hypophyses. Spayed rats are only 7.14% heavier and the difference is not significant. But if one compares the relative weights in different groups of animals, an astonishing relationship between hypophysis and body weight will be seen. The female hypophyses attain a weight which is both actually and relatively greater than in males. Moreover, from comparisons between gonadectomised and control animals, it can be seen that the castrated rats grew larger hypophyses than normals, whereas in females, the spayed did just reach the weight of control females. This seems to show that there is a sex difference in response to gonadectomy so far as the hypophysis is concerned. The castrated rats failed to gain as much body weight as the normal males, yet they grew a larger hypophysis. The spayed females grew faster than their controls, yet they have a relatively smaller hypophysis.

Hatai (1913) using 25 castrated rats to compare with 12 controls, reported that the average body weight of castrates was 122 grams and of controls, 123.5 grams at the same age. He found a striking difference in the weight of hypophysis between castrated and control rats. This difference amounted to 73.62% in favour of the castrated. In female rats, the spayed gained 28.4% more body weight than controls, yet the hypophysis was only 3.84% greater in weight. Korenchevsky (1934) and Lawless (1936) also found hypertrophy of the hypophysis in castrated rats. Freudenberger and Howard (1937) and Freudenberger and Hashimoto (1937) reported significant increase in the hypophysis of spayed rats, but Freudenberger and Billeter (1935) did not find a significant difference. Moore (1922) working with guinea pigs, observed that normal male hypophysis is 14% heavier than that of the castrated. When the relative weight of hypophysis was considered, the normals were still 7.6% greater than test animals. In female guinea pigs, the spayed had a 1% greater body weight than controls, yet their hypophysis was 12% lighter than the latter. Moore claimed that the comparison of animals from the same litter does not offer any advantages in favour of problems of this type, and he did not mention how he selected the 12 normal and 11 castrated males, and the 12 control and 11 spayed female guinea pigs used in his experiments. The fairness of his comparisons therefore remains doubtful.

By far most investigators are agreed that hypertrophy of the hypophysis follows the removal of the testes. This effect will be further discussed later.

Eyeballs:- A significant difference, i.e. 21,3%, is shown between male and female controls, but it is decreased to 13.76% between test rats. Test males are 2.51% smaller, test females 7.2% greater in their weight of eyeballs than their respective controls, yet they are insignificant. Freudenberger and co-workers also failed to see any significant difference between spayed and normal rats.

Submaxillary gland:- On the average control males have a 22.4% greater submaxillary gland weight than control females. Between test males and females the difference is 12.05%. No significant difference is found between treated and control animals within their own sexes. Freudenberger and Hashimoto (1937) found a significantly increased weight in spayed females, but Freudenberger and Billeter (1935) found no significant change.

Thyroid gland:- Though the control males have a 9.6% greater weight for their thyroid gland than control females, yet it is not significant owing to the wide variations existing. The difference between test males and females, 11.9%, is insignificant. Test males have a 1.8% greater and spayed 1.9% smaller weight than their controls, but both are insignificant. Freudenberger and associates working with females rats found a significant change after spaying in one of their experiments, a probable one in another, yet no change was noticed in a third. The results from their laboratory indicated, they state, that there is an early hypertrophy of thyroid which later disappears. This was said to be also true of the hypophysis.

Hatai (1915) was bewildered at the wide variations of the thyroid gland. For instance, the deviation of thyroid weight in castrated rats from expected values ranged from -0.8% to -90.3%, the average being 21.5% less than that in control males. In spayed females, he found only three out of six groups which showed an increase, the remainder showing a decrease, with an average of 3.3% excess over controls despite the fact that the former have a much greater body weight. Moore (1922) found in normal guinea pigs a 3.2% greater weight of thyroid in females than in males. In gonadectomised animals, the thyroid of castrated males gained 8.6% more, and spayed females 6.8% more over their respective controls. The variation as shown from actual weight was also very great.

A recent paper of some interest in this connection is that of Todd, Wharton and Todd (1938), who thyroidectomised one of each of five pairs of twin lambs. Their study of the maturation of the skeleton, particularly of the epiphyseal union showed that the removal of the thyroid gland reduced the velocity of growth of the shafts of the bones and resulted in a defective maturation, particularly of the epiphyses. In spite of the lack of definite results in the present experiment, castration may affect mature skeletal size through a modification of thyroid activity.

Thymus:- Castrated male rats showed an increase in thymus weight of 46.3% over that of controls; the spayed increased 31.3% more than normal females. The thymus weight of female controls was found 3.9% greater than that of male controls, but after spaying the females are 6.75% lighter in their thymus than castrated males.

So far as the present writer is aware, the findings of numerous investigators fully agree upon the fact of the enlargement of thymus caused by gonadectomy. Since involution of thymus closely corresponds with time of puberty, it is conceivable that as soon as the gonads are removed this natural process will henceforth be postponed. Donaldson (1924) prepared a chart showing that the weight of thymus in albino rats grows on a very steep line up to about 80 to 85 days of age, then it declines with the advance of age, though the descending line is much flatter than the ascending one. The thymus is not altogether a lymphoid organ, but is also concerned with nutrition, growth, and balance of other endocrine activities. Although the greater weight of thymus in gonadectomised animals is universally admitted, yet there is one point in dispute among investigators -- some maintain that the thymus is merely persistent after gonadectomy, while others actually observed a great growth response of atrophied thymus.

Hatai (1915) reports that the thymus of castrates is on the average 61.2% greater than that of the controls. Spayed females showed an average increase 58.3% as comparedwith normal females. In mice Masui and Tamura (1926) found that the thymus weight of normal females is considerably heavier than normal male mice,i.e. the difference being on the average 81%. Castrated mice had an excess of weight of 146% over controls. This difference is even greater than that between control males and females. Spayed females only grew 3.7% greater than controls, yet the body is considerably heavier for the former. They concluded that castration is followed by a striking increase in weight of thymus which approaches that of the female thymus, but that spaying has little effect. The present data, secured from rats of 100 to 120 days of age supports the view that gonadectomy increases the relative weight of thymus in both sexes.

Heart:- The heart weight in male controls is 28.97% greater than female controls, while the difference between test animals is 17.14%. Castrated rats are 4.5% lighter in their heart weight than normal, and spayed 11.37% greater. Lawless (1936) also noted a significantly smaller heart in castrated rats. Freudenberger and associates found a significant difference in two investigations, a probable one in another. Relative to body weight, heart weight does not appear to be much affected by gonadectomy.

Lungs:- Lungs of control males are significantly heavier than those of control females, but no significant changes can be detected from comparisons of other groups. Lung weight is most variable, the coefficient of variability ranging from 17 to 29%. It is not known how far this variability may be attributed to variation in amount of contained blood.

Liver:- A very significant difference is shown in the weight of liver beteen control males and females, being 28.9% in favour of the former. For some reason the liver is relatively lighter in gonadectomised animals.

Spleen:- Moore (1922) noted that spleen weight for normal female guinea pigs is 6.6% greater than normal males. After gonadectomy in both sexes a deficiency was observed. Masui and Tamura (1926) also reported a decreased weight for gonadectomised mice. However, Freudenberger and associates (1935, 1937, 1937) working with mice found greater weight for spayed females in all three series of experiments. In the present investigation, a difference of 18.21% was found between control males and females favouring the former. Among the test animals the difference becomes 15.19%. An increase of 5.14% for castrated males and 1.39% for spayed females was indicated from a comparison with their respective controls. As the spleen is a very variable organ, these comparisons mean very little. It seems however safe to conclude that males have heavier spleens than females.

Suprarenal glands:- Suprarenal gland weight of female rats is strikingly greater than that of males, in spite of the fact that the latter are greater in their body weight. This difference is 57.4% in favour of control females, and 19.5% for spayed females. Castrated gained 16.6% over normals, while spayed females showed a decrease of 11.4% compared with controls. These are all significant. Hatai (1915) reported a characteristic alteration after gonadectomy. He found that the suprarenal weight of castrated rats exhibited an increase of 8.5% to 17.6% on the average, while those of the spayed rats show a decrease of 5.3% to 25% compared with their controls. Investigations on the effects of ovariectomy made by Freudenberger and Billeter (1935) and Freudenberger and Howard (1937) found a significant decrease of suprarenal weight in spayed rats, but Freudenberger and Hashimoto (1937) did not find significant change in their series; the animals used by the last mentioned authors were closely comparable to those of the first. In castrated rats Korenchevsky (1930), Anderson and Kennedy (1933) and Lawless (1936) all reported greater weight for suprarenals. In guinea pigs, Moore (1922) noted that normal male suprarenal weight is 20% greater than those of the normal females, the average of the body weight of the two sexes being almost identical. Castrated males showed a decrease of 7.6% as compared with controls, spayed females became 22.7% smaller. It may be concluded that in rats males have larger suprarenal glands than females, castration increases and spaying decreases suprarenal weight.

Kidney:- Kidney weight of male controls is 25.9% greater than that of female controls, but it is reduced to only 5.9% in test animals. Comparing male controls with castrates a difference of 18.6% was observed favouring the former, while in females it was 3.1% in favour of the spayed. Masui and Tamura (1926), Korenchevsky (1930) and Lawless (1936) all find the same results from castration, and Freudenberger and co-workers from spaying. It would be interesting to know why castration should affect the kidney weight so markedly.

Stomach:- The weight of stomach between test and controls can entirely be attributed to the difference in body weight. Although the weight of stomach with contents was recorded, it does not bear much significance in itself as the amount of food stored in the stomach and intestines may vary considerably between individuals and from time to time.

Intestines:- The weight of intestines of male controls is only 9.7% over that of the females; however, the weight of empty intestines is 21.4% greater and should be considered more reliable as has been explained above. Again, female test rats had 0.8% heavier intestines with contents, but empty intestines were 11.9% smaller. Weight of intestines with and without contents for male controls are 5.6% and 5.3% greater than test males respectively; obviously they are closely correlated. In spayed females, these become 5.4% and 6.2% greater than controls. Freudenberger and associates found the same results. It is clearly indicated that the weight of stomach and intestines are closely associated with the size of the animal. The larger size of an animal may depend on a larger capacity for food consumption and utilization; equally well the relation may be inverted, or both may depend on a growth or size factor.

Fat:- No attempt was made to extract all the fat out of the body with ether or alcohol. Only the gross visible fat from the renal and inguinal regions, as well as subcutaneous fat was weighed. The absolute fat weight of control males is 4.2% greater than control females, nevertheless, when body weight is considered, the female controls have greater relative fat weight. Spayed rats had 12.6% more fat than test males. An increase of 4.2% was found in castrates as compared with control males. The spayed females gained 22.53% more. But none of these differences are significant owing to the great variations. Nevertheless, it may be suspected that spaying does tend to increase the amount of fat.

Male accessory genital organs:- It is to be expected that the accessory reproductive organs of male test rats should fail to develop as soon as the primary sex organs were removed. The male accessories weight in castrates is only 4.4% of that of the control male rats.

Uterus:- Spayed female rats only had an uterus weight 14.1% of that of normal females, showing once more that the development of the uterus is conditioned by the activity of the ovaries.

5. Bone growth:- An attempt was made to determine whether the skeletal growth was affected by the removal of the sex glands. Bones from eight litters of animals were used, including 15 normal males, 14 castrated males, 16 female controls and 16 spayed females.

(1) Measurement of tibia and hip bone length:- The length of tibia and hip bone are given in Tables 11 and 12. Figures 10 and 11 give an illus-

tration of the differences in bone length between test and control animals.

On the average the normal male controls have a 2% greater tibia length and 5% greater hip bone length than castrated littermates. On the other hand, the spayed females have an excess of tibia length of 2.6% and of hip bone length of 1.3%. These differences are not very large, but seem to indicate that the greater body weight of control males than of castrates and spayed females than of controls is based in part at least on greater skeletal development Figures 10 and 11 show the actual photographs of tibiae and hip bones of gonadectomised and control rats.

(2) Examination of epiphyseal union:- X-ray photographs of live rats have been taken using five litters of animals, with the intention of showing up any difference in skeletal growth. The results were negative, no significant differences could be recognised from the plates. Examination of the stage of epiphyseal union was also made. In human and some other species it has been reported on various occasions that the removal of testes causes delay or cessation in the union of the epiphyses, thus

resulting in longer bone growth. Apparently the animals used in the present investigation were sacrificed too young, for no differences were detected either between test and control rats or between males and females. Epiphyseal union had already taken place at: (1) capitellum and trochlea; (2) Proximal radius; (3) distal tibia; (4) tuberosity of calcaneum; (5) distal fibula; (6) head of scapula; (7) hip bone (primary elements). Epiphyses not united were: (1) medial epicondyle of humerus; (2) olecranon of ulna; (3) head of femur; (4) greater trochanter; (5) lesser trochanter; (6) distal femur; (7) proximal tibia; (8) proximal fibula; (9) distal radius; (10) distal ulna; (11) head of humerus; (12) base of coracoid.

Table 11

Length of tibia and hip bone of male rats in cm.

Litter no.	Tibia Rat no.	Contr.	Rat no.	Castr.	Hip bone Rat no.	Contr.	Rat no.	Castr.
1	42	3.65	41	3.5	42	3.95	41	3.73
	44	3.55	43	3.45	44	3.85	43	3.7
2	65	3.8	63	3.6	65	4.1	63	3.8
	66	3.8	64	3.7	66	4.2	64	3.95
3	74	3.55	73	3.58	74	3.8	73	3.75
	76	3.65	75	3.75	76	3.9	75	3.8
4	80	3.6	79	3.5	80	3.93	79	3.65
	82	3.65	81	3.6	82	3.95	81	3.8
	84	3.63	83	3.7	84	3.95	83	3.95
5	93	3.45	92	3.6	93	4.0	92	3.75
			94	3.4			94	3.7
6	109	3.7	110	3.5	109	4.05	110	3.75
	111	3.75			111	4.2		
7	116	3.65	115	3.55	116	4.1	115	3.75
	118	3.4	117	3.5	118	3.6	117	3.85
	123	3.8			123	4.1		
Mean		3.642		3.566		3.978		3.780
Mean diff. in % of control			2.09				4.98	

Table 12

Length of tibia and hip bone in female rats in cm.

Litter no.	Tibia Rat no.	Contr.	Rat no.	Castr.	Hip bone Rat no.	Contr.	Rat no.	Castr.
1	46	3.3	45	3.45	46	3.6	45	3.6
	48	3.25	47	3.4	48	3.5	47	3.63
2	69	3.4	67	3.5	69	3.73	67	3.7
	70	3.43	68	3.45	70	3.7	68	3.7
3	78	3.3	77	3.3	78	3.53	77	3.5
4	87	3.28	89	3.45	87	3.7	89	3.75
	88	3.3	91	3.5	88	3.53	91	3.7
	90	3.33			90	3.7		
5	95	3.35	96	3.3	95	3.6	96	3.7
	97	3.35	98	3.15	97	3.63	98	3.5
6	113	3.5	112	3.4	113	3.7	112	3.7
			114	3.4			114	3.8
7	119	3.25	121	3.3	119	3.6	121	3.5
	120	3.2			120	3.5		
	122	3.15			122	3.15		
8	125	3.3	124	3.38	125	3.65	124	3.7
	126	3.3	127	3.53	126	3.65	127	3.75
			128	3.48			128	3.73
			129	3.4			129	3.6
Mean		3.31		3.40		3.61		3.66
Diff. of means as % of control			-2.63				-1.27	

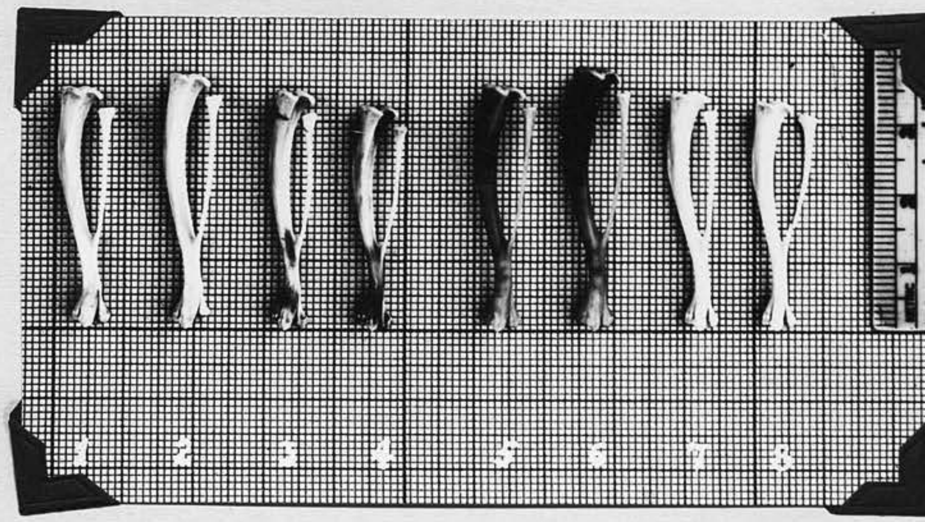


Fig. 10. Tibiae and fibulae from control and gonadectomized rats (see below)

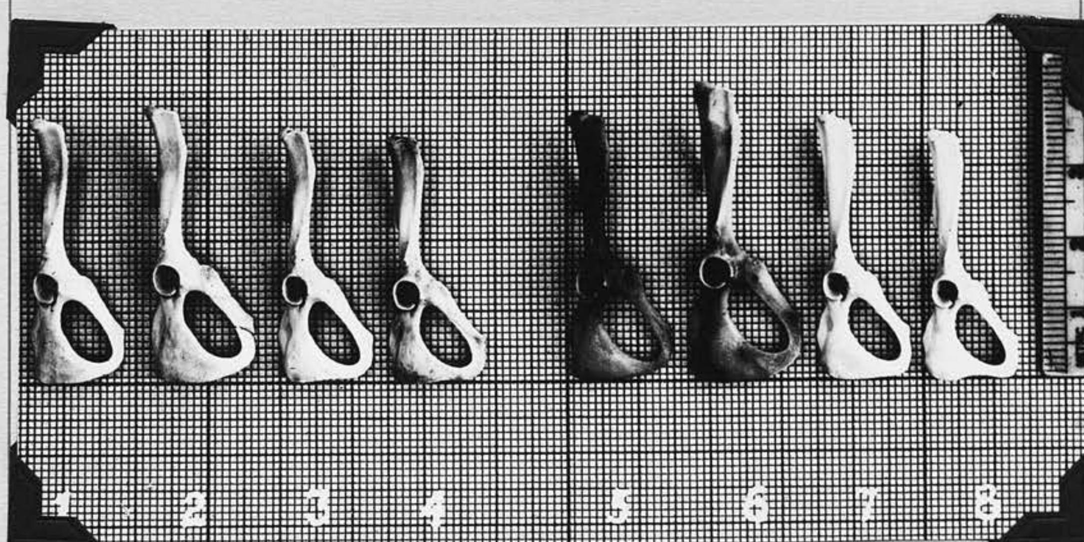


Fig. 11. Hip bones from control and gonadectomized rats.

No. 1, 5, Castrated males (rat nos. 43, 110)
2, 6, Control males (rat nos. 42, 111)
3, 7, Spayed females (rat nos. 45, 89)
4, 8, Control females (rat nos. 48, 88)

DISCUSSION.

Growth rate:- In the growth curves of control and gonadectomised rats (Fig. 1 and 2,p.33-34), the reduction of sex difference in operated animals is clearly illustrated. The decreased growth rate in male castrates and increased rate in spayed females indicate an intermediate type of growth curve.

Evans and Simpson (1927, 1931) induced artificial gigantism in rats by injection of anterior hypophyseal extract. They found that the greatest contrast of gonadectomised rats with controls is exhibited by injected spayed females, and least by injected normal males. The males were not only incapable in general of adding to their normal body size to the degree which characterises females, but were also more variable than the latter.

It has been revealed from genetic and cytological studies that sex is determined by sex chromosomes which are but a small part of the total chromosome complement in large animals. Within a certain period following fertilisation which is specific for each species, embryoes begin to develop in a way characteristic of a particular sex. But there are instances in which they fail to accomplish the sex difference properly and come to resemble both sexes or to be intermediate between them. This indicates that any organism can be either a male or a female depending on the sex-determing mechanism which merely guides the developing organism in the direction of one sex or the other. When the mechanism fails, development does not stop but proceeds in a way not normal for either sex. Therefore it is not surprising that after gonadectomy, both sexes will tend to be intermediate between the two, although from genetic point of view this is not a necessary consequence. It is true in fact because the presence of ovaries retards female growth in rats as measured by gain in weight, whereas the presence of testes in a genetically similar animal, if it affects growth at all, accelerates it.

Since gonadectomy can only be performed after birth, characteristic sex differences have already developed to a certain extent. Though they may be minimised by preventing further development of the reproductive organs, they can not be entirely eliminated.

Compensation is a well known phenomenon in living organisms. Hatai (1915) demonstrated that the compensatory growth of the remaining ovary of semi-spayed rats is almost perfect and thus the single ovary attains twice its normal weight, though the remaining testis in semi-castrated males only increased 14 per cent of its normal weight. The overgrowth of females after ovariectomy and the greater response to growth-stimulating hormone administration is perhaps another phase of compensation. The size of an animal is mainly determined by certain genes, but it is also subject to many other factors. Nutrition, temperature, light, humidity, etc. all affect ultimate size, but the endocrine glands, especially the gonads, have also an important influence. If all other endocrine organs, such as hypophysis and thyroid glands, function normally, an animal will grow to a size characteristic of its particular sex. A male and a female may be very similar in genotype so far as size-determing factors are concerned, but their growth is conditioned by sex. Since a female is handicaped in its growth by the presence of ovaries, it still possesses the hereditary capacity to grow as soon as the ovaries are removed.

Growth response can also be evoked after retardation by other methods. The body weight of rats was held by Jackson (1936) at a nearly constant weight (about 50 grams) for 15 weeks by a protein-deficient diet. When refed on normal stock diet, the females rats had overtaken at nine months of age the normal controls in average body weight. The males, however, still lagged significantly behind their controls at one year of age. In another experiment, Jackson (1937) again observed abnormally rapid growth of underfed rats after return to full feeding. These examples of compensatory growth can be interpreted as due to the inherited character of size. Modifications may be made by many internal or external environmental factors, but whenever there is a chance to escape from such influence, animals tend to show some form of compensatory growth.

Efficiency:- The most striking and significant difference in the efficiency of food utilization as calculated by the efficiency quotient adopted is found between control and spayed females, being 30.96% in favour of the later. The spayed females in Group A had a 13.5% and in Group B 8% greater body weight than their respective controls, yet the food intake was on the average 2.9% less. The increased growth rate of the spayed females has therefore to be attributed mainly to higher efficiency and not to increased food consumption.

Slonaker (1930) reported that the daily food intake in calories at the age of 200 days for the male rats was: normals, 101.28; vasectomised, 54.7; castrated, 55.4; for females: normals, 74.5; hysterectomised, 67.9; ovariectomised, 45.2. The percentage of available energy for growth and basal metabolism at the same age for the males was: normals, 27.5; vasectomised, 78.9; castrated, 83.3; for females, normals, 26.9; hysterectomised, 34.5; ovariectomised, 84.2. The growth of the different groups is correlated with the amount of energy available for growth and basal metabolism.

Holt et al (1936) stated that the food intake per gram of body weight was approximately the same for castrated and control male rats. The amount of food eaten by spayed females was more than by control females, but the food intake per gram of body weight was less. From these reports it may be concluded that spayed females are decidedly more efficient than normal females, at least in albino rats. Experiments on different kinds of farm animals to test the usefulness and practibility of spaying from this aspect might be worthwhile.

The castrated male rats are less efficient than their controls. This is due to slower rate of growth rather than difference in food consumption. However, in view of the fact that studies on the growth of castrated animals are conflicting, far-reaching conclusions in this respect can hardly be drawn.

The E.Q. values of test and control males is higher than for test and control females. This difference may chiefly be attributed to the slow growth rate of female rats. But the greater spontaneous activity and the relatively larger surface area, which cause larger energy expenditure,for maintenance, probably affects the efficiency of females.

Body size and proportions:- Female rats grow to a heavier body weight after ovariectomy. This is almost universally recognised. The elimination of the effects of oestrin suggests itself as the cause. From the results reported by Freudenberger and Clausen (1937) the inhibitive effect of oestrin on body weight can be postulated. They injected 25 rats of three weeks old with a dose of 200 I.U. every other day for eleven weeks. From the fourth after the treatment the test animals fell behind the controls in weight. After the fourteenth day the difference became significant. At the end of the experiment the test rats had a 20 per cent smaller weight than controls. Accepting this theory regarding the influence of oestrin as an explanation for the time being, one may ask whether any information about the mode of action of the hormone can be obtained from the reaction of the various kinds of tissues to its removal. It has to be remembered that the reaction of the tissues may depend on the stage of development which they have reached. Also, there may be genetic differences in the reactivity of the tissues. Further, the uncontrollable variability of some of the organs makes it more or less impossible to detect slight changes in them.

In spite of these limitations, several general principles seem to emerge. The removal of the ovaries results in increased growth of both skeletal and muscular tissue and therefore does not change the natural conformation of the rats very much. It is possible that if the rats had been dissected when they were older, they would have been more plump with subcutaneous and internal deposits of fat. Bone growth, however, seems to be but little affected. Yet it should be pointed out that in sheep and cattle castration does affect both bone length and thickness(Hammond 1932).

Gonadectomy is generally supposed to result in the accumulation of more fat in both sexes and in the present data evidence in support of this can be found. However, it is not possible to tell whether this is due to the absence of a direct effect of hormone on fatty tissues, or whether it is due to lessened activity and a larger proportion of food energy available for growth, In the present state of knowledge the latter seems the more likely.

The effects of gonadectomy on the liver and kidneys have not received much attention but the changes in these organs following castration are so marked as to deserve more mention. In castrates the liver was distinctly smaller and the kidneys significantly larger both absolutely and relatively. In the females, the effects of ovariectomy are not so clearly marked, and it is doubtful whether these organs were affected in a particular way.

The general effects of gonadectomy do not apply to the system of internal secretion. The changes taken place in them require individual examination.

Hypophysis:- Before proceeding to deal with the hypophysis as a whole, one report on the effect of the growth-stimulating principle of the anterior lobe of the pituitary body on growth and food utilization may be cited. Nelson, Palmer and Kennedy (1934) injected rats with growth hormone preparations. A few of the animals were treated with Phyone and the remainder with antuitrin G, about 1cc. daily. 57 male and 66 female rats, with 49 male and 51 female controls were used. It was found that growth and the efficiency of food utilization were apparently increased by such treatment within certain limits of heredity and sex. The females responded to the injection much more markedly than males,

It has been shown in the preceding pages that both gonadectomised males and females attained greater hypophysis weight than their respective controls. But considering the significant difference in body weight between test and control animals, a direct comparison of the observed weights does not seem to be fair. The castrated males have a greater relative hypophysis weight than controls, whereas the weight of hypophysis in females in respect to body weight is the same after ovariectomy. Or put into other words, a decreased body weight following gonadectomy is accompanied by an hypertrophy of hypophysis, and increased body weight by no change in relative hypophysis weight.

Hatai (1915) also noticed that the lack of hypertrophy of hypophysis in spayed females is consistently associated with overgrowth and obesity. Castration in males produced a striking enlargement of hypophysis, but overgrowth and obesity are absent. This led him to believe that the product of the unaltered gland in spayed females must be employed for two purposes; namely, to replace the ovarian hormone and for the normal uses whatever these might be. These phenomena were stated to be similar to those following hypo-secretion of the hypophysis. The compensatory hypertrophy of hypophysis in castrated males appeared to prevent hypo-secretion, and therefore, overgrowth and obesity were also prevented. But in view of the well-established fact that hypo-secretion of the pituitary results in infantalism, and that hyper-secretion will bring about gigantism or acromegaly, Hatai's hypothesis seems to be hardly justified. It is true that the pituitary body regulates ovarian activity, but in the absence of the ovaries, with the degeneration of the whole reproductive system, no such function can be performed.

Livingston (1916) conducted two series of experiments on the effect of gonadectomy in rabbits on the weight of pituitary, comprising 60 animals in the first series and 90 in the second. He could not find a constant sex difference in hypophysis weight in normal animals, nor constant hypertrophy of hypophysis following gonadectomy. Yet he has noticed that in those animals which show an increase in body weight there was a lesser hypertrophy of hypophysis, whereas in groups where no effect can be shown upon the growth rate a distinct hypophyseal hypertrophy is constant though not very marked.

Freudenberger and Clausen (1937) advanced a suggestion to account for the smaller body weight of female rats after oestrin administration. The initial effect may be to stimulate the hypophysis, causing the liberation of increased amounts of thyreotropic, gonadotropic and adrenotropic hormones. The secretion of hypophyseal hormones decreased later due to former over-activity of the gland. This resulted in a smaller animal body. However, they admit that there is no initial increase in the amount of growth-stimulating hormone, or if there is an increase in amount of this hormone, its effects are neutralised in some manner.

With our present incomplete knowledge on the relationship between endocrine function and growth of an animal, we are not yet in the position to interprete the modified growth of the body and hypophysis following gonadectomy. But certain facts may be recalled. Firstly, the association of a decreased body weight and hypertrophy of hypophysis, either produced through castration or by oestrin injection, has been agreed on by many investigators, though there are few exceptions (Moore 1922), Secondly, the potency of the hypophysis hormones is not necessarily related to gland size. As has been shown by Evans and Simpson(1929), the pituitary of immature male rats is twice as potent as the much larger glands of adult females. The adult male pituitary is about three and one half times as potent as that of the female. Therefore, we need to determine the potency of the hypophysis rather than merely to measure its size. Thirdly, the disturbance of the normal physiological activities in the animal body after the removal of gonads may indirectly or directly affect endocrine glands other than the pituitary, and in attempts to find a solution, they should not be neglected, especially those concerned with metabolism.

Thyroid gland:- The normal variability in the weight of thyroid is surprisingly great, which renders it difficult to attempt an interpretation of the data available from the present experiment. However, since it is one of the chief endocrine organs with a great influence on metabolism, its changes resulting from gonadectomy can not be disregarded.

Hatai (1915) stated that while other endocrine glands all showed their sex differences in size, the thyroid gland was without sex difference. Livingston (1916) observed a moderate decrease of thyroid weight in castrated rabbits. Korenchevsky (1930) noted a 30 per cent lighter average weight of thyroid in castrated rats, but he did not draw conclusions owing no doubt to the wide variations. Lawless (1936) found slight decrease in thyroid weight in most of the castrated rats.

Data obtained from the present study regarding thyroid gland weight do not show much difference between test and control rats. No effect of gonadectomy on its size can be claimed, but whether the intensity of its function ~~whether~~ has been altered is doubtful.

Thymus:- The thymus weight is significantly increased in the gonadectomised male and female rats over their controls. It may not due to antagonism between gonads and thymus, but the development of the reproductive organs appears not favourable to the thymus. Freudenberger and Clausen (1937) observed that the thymus became smaller in oestrin-injected female rats before the body weight decreased. This led them to suspect the possibility that the thymus might be responsible for the smaller body weight in treated animals. This supposition does not appear to allow for involution of thymus in normal growing animals.

Suprarenals:- The decreased body weight of castrated males is associated with a significant increase in suprarenal weight, whereas the spayed females show a smaller suprarenal weight than normal female rats. Since this gland is concerned with both sexual activity and metabolism, its differential response to the removal of testes in males and ovaries in females can not be overlooked.

The greater suprarenal weight is exhibited in both groups of castrated males. The difference reached was more pronounced in those animals castrated at an older age, the difference of Group B amounted to 55.9%, more than three times greater than that between Group A males. Group A spayed females have a suprarenal weight about ten per cent less than that of controls; the difference becomes 14.6% in Group B females. The cause of such a distinct sex diffrence in response to gonadectomy is not understood.

An interesting feature of the response of the suprarenals to gonadectomy is its resemblance to that of the hypophysis. In both cases, the glands became larger following castration. After spaying, the suprarenals became smaller, while in the hypophysis no change in relative weight was found, although others have found it. This similarity in response suggests that their functions are linked.

Accessary reproductive organs:- The hypotrophy of the male accessary organs in castrated rats is almost complete. The organs affected are the ductus deferens, the prostate gland, the seminal vesibles and the coagulating glands attached to them. The failure of development of these organs has been shown to be due to the absence of testes hormone. Modern methods of assay of hormone from testes utilised this fact. Potency of the hormone can be determined by the amount of reaction of the accessory organs to testes hormone injection.

Uterus in spayed females underwent the same fate as the male accessaries.

SUMMARY

1. An experiment has been carried out to determine the influence of gonadectomy on the growth of body and organs, and on the efficiency of food utilization of albino rats. The material comprised 16 castrated males, 18 control males, 20 spayed females, and 21 control females, a total of 75 rats.(Table 1, p.19).

2. Beginning at 50 days of age the rats were fed and housed individually for a period of 10 weeks. Body weight and food consumption were recorded twice weekly. A special food container was devised to provide reliable measurements of food consumption (p.20).

3. Gonadectomy took place at three ages, 1, 14, and 45 days, but the rats operated on at 1 and 14 days have been considered together. Incisions were made at the same time on the control rats.(p.23).

4. At the end of the experimental feeding period, all the rats were autopsied. Besides total body weight and length, the weights of all the principal organs, of the endocrine glands, and of fat were recorded.(p.25).

5. As a measure of economy of food utilization, Efficiency Quotients have been calculated from the formula :

$$E.Q. = \frac{\text{Gain (gr.)}}{\text{food (gr.)}} \cdot \frac{(\text{init.wt. plus } \frac{1}{2} \text{ gain})}{100}$$

6. The effect of gonadectomy on the gain in weight of the females was to increase it by 12% over the female controls, while in the males it decreased it by 8.8% (Table 3, p. 30).

7. The food consumption of the gonadectomized rats was less than that of the controls (Table 4, p.37). The mean values of the E.Q. were found to be :

Control males	0.220	Control females	0.125
Castrated males	0.196	Spayed females	0.155

The lower values for females generally are attributed to their greater bodily activity. Spaying reduces this activity and thus raises the E.Q.(Tables 5 and 6, pp. 44, 45).

8. A tendency was observed for the E.Q. to diminish from the beginning to the end of the experiment, probably as a result of the greater maintenance needs of the rats.

9. Comparison of carcass measurements on the males showed significant differences only in respect of total body weight, and weights of hypophysis, thymus, liver, kidneys, suprarenals, and male accessory sex organs. (Table 8, p. 49).

10. Similar measurements on the females (Table 9, p.50) showed significant differences in more respects but not in weight of hypophysis, liver, or kidneys. The thymus was significantly larger, and the suprarenals significantly smaller in the spayed females than in their controls.

11. Coefficients of variability have been calculated for each organ weight (Tables 8, 9, pp. 49,50). The findings are in agreement with previous work. There were some very variable organs including lungs, spleen, fat, genital organs, thymus, and suprarenals. Other organs were of medium variability. Total body weight and nose-anus length were of low variability (p.61).

12. The weight of hypophysis was actually and relatively larger in the castrates than in the controls. In the spayed females on the other hand, it was actually larger but relatively the same weight as in the controls. (Tables 8, 9, pp. 49,50).

13. The thyroid gland was found to be so variable that only a large difference following treatment could hope to be established, but actually the differences between the groups were very small.

14. Considerable changes were observed in thymus weight following gonadectomy. Castrates showed an increase of 46.3% in weight, and spayed females an increase of 31.3%.

15. The suprarenal glands were relatively much larger in the castrates than in the control males. On the other hand, they were relatively much smaller in the spayed females than in their controls. There was thus a certain degree of parallelism in the response of suprarenals and hypophysis to gonadectomy.

16. All accessory male sex organs and the uterus in females became almost completely atrophied after gonadectomy.

17. Kidney weight was significantly greater in the castrated males-- the difference amounted to 18.6% of the control weight. No change occurred after spaying.

18. The lengths of tibiae and hip bones from 8 litters have been recorded (Tables 10,11, pp. 78, 79), and although not significant, the differences observed suggest that skeletal development is influenced by gonadectomy in the same direction as body weight up to the age at which the rats were killed (p.76).

PART II

A PRELIMINARY REPORT ON THE EFFECT OF SELECTION FOR EFFICIENCY OF FOOD UTILIZATION IN RATS.

INTRODUCTION.

The introduction to British Agriculture of the turnip began an improvement in stock feeding which has now come to the point where further advances are becoming difficult to make. During this improvement the art of the stockbreeder flourished and the foundations for the modern breeds were laid down. The principal problem of livestock breeding was to provide animals which were capable of taking advantage of the better husbandry conditions to produce more meat and milk which were being demanded in increased quantity and higher quality. The success of these efforts has been considerable, but the rate of progress appears too slow by modern standards. Many attempts are therefore being made to improve on the methods of the past and to develop new ones.

The essential fact of animal breeding is the selection of some animals in preference to others for the production of the next generation. As might be expected breeders used as a basis for selection those characteristics of animals which could be readily judged, namely, growth rate, conformation and productivity in terms of milk, fertility and so on. The average farm animals of today are not yet perfect in these respects, but many breeders think that it is time that more attention was paid to important characters such as disease resistance, constitution, and efficiency which are not easily measured.

One of the most specialized farm animals is the dairy cow, and it is from experience with her that much interest in efficiency has arisen. When milk recording started much importance was placed in total lactation yields and led to the production of some remarkably high performances. It was discovered however that in many cases the very high producing cow did not breed reliably or cost more feed than the milk was worth. Many investigations have since shown that among dairy cattle and pigs there are large differences in the amount of food consumed in relation to the amount of milk or bacon produced. For economic reasons therefore it is desirable to breed strains of domestic animals which are as economical in their use of food as possible consistent with their purpose. When attempts are made to improve breeds by selection for efficiency of gain there are many questions arising. In the first place it is necessary to know whether efficiency is a character in the genetic sense, and whether it shows genetic variation which can be separated from variation due to environmental causes. Secondly, it is desirable to know what influence such factors as sex, age, rate of growth, plane of nutrition and composition of food stuffs have on efficiency so that animals differing in these respects can be compared.

Numerous investigations have been reported on the efficiency of the different feeding-stuffs for growth and maintenance of animals. Conclusions are usually drawn from comparisons of growth and state of health between test and control animals, on the assumption that if other environmental factors are controlled, the difference in growth rate can be attributed to the effect of the nutrients supplied. Growth, however, is not such a simple matter. As will be shown below, for instance, growth rate and efficiency are closely interdependent and food stuffs which reduce rate of growth may affect efficiency only indirectly. Further, it is known that ultimate size is governed by the size-determining genes carried by the organism, and it seems highly possible that the process of growth is related to ultimate size. McPhee and Eaton (1931) in a study of the genetic growth differentiation in guinea pigs stated that some of the inbred lines showed considerable difference in the age at which a certain percentage of the mature weight is reached. The fast-maturing animal was usually much smaller in ultimate size than the slow-maturing one. Todd et al (1938) suggest too that heritable differences in thyroid activity affect growth rate and ultimate skeletal size in sheep. Therefore, the importance of heredity on growth should always be kept in mind when attempting the interpretation of results from a feeding experiment.

At present it is very difficult to recognise genetic differences in efficiency among similar animals, not only because the many environmental conditions complicate the situation, but alsodue to the fact that the expression of such a character as efficiency is not clear cut; and like many other cases of multiple factor inheritance the factors may have additive effects or may interact in quite other ways. Since a direct approach to the problem of the inheritance of efficiency cannot as yet be made, indirect methods must be adopted, such as the study of the effect of changes in environment, and of selection based on the best criteria available. The purpose of the present study was to obtain information regarding the effects of selection and inbreeding on the attainment of a more efficient strain of rats. Owing to limited time the data so far collected do not provide complete answers to the questions involved. Several interesting facts have emerged however which will be described. Discussion will be limited to consideration of the efficiency in relation to growth rate and food consumption.

REVIEW OF LITERATURE.

The classic work on the effect of inbreeding on the growth of body weight was conducted by King (1918) at the Wistar Institute. In order to test the validity of the theory held by Charles Darwin and his contemporaries that inbreeding invariably resulted in a loss of size, constitutional vigor and fertility, she started with two pairs of littermate rats and inbred them by brother-sister matings for fifteen generations. Records of 333 males and 306 females were collected. She found that the inbred males and females of the fifteenth generation were about 40 per cent less variable than the controls. Males from the 7th to 15th generations were heavier than stock rats. The adult of the inbred males was on the average 18% greater than stock animals. Females at one year of age were 3.7% heavier than controls. King (1919) continued the experiment to the 25th generation. The extra 296 males and 310 females confirmed her previous findings. There were instances of smaller body weight or higher variability as time went on but on the whole the inbred rats were superior to the control reared under the same conditions. Inbreeding practised with selection did not bring about deterioration but on the contrary improved the stock. This extensive experiment did much to shake the prejudice against inbreeding prevalent at that time. However, as the author stated that the animals were not always well fed and were sometimes even subject to malnutrition, some reserve has to be maintained regarding the results.

McPhee and Eaton (1931) reported an investigation on inbreeding of guinea pigs carried out at the laboratory of the Bureau of Animal Industry, U. S. D. A. for twenty-five years (1906-1930). Five families were bred by sib-mating and a family of controls, maintained by breeding only from animals more distantly related than third cousins. The variations in adult weight were lower among the inbred lines than the controls by about 40 per cent. The growth rate and the age at maturity differed among families. The greater the mature weight, they stated, the longer is the time required to reach a given percentage of this weight. Marked increase in growth and weight were obtained by crossing lightweight families. Crossing between heavyweight and lightweight families resulted in growth curves similar to the heavyweight family. Crosses between two heavyweight families did not improve growth. This experiment was further discussed by Eaton (1932) who stated that there had been a decline in all measures of vigour, but decline in fertility had been the greatest and in growth the least, but he explained that inbreeding was apparently not responsible for much of the decline in the inbred stock, for the control stock had suffered almost a parallel decline, though at a higher level. Nevertheless, in view of the fact that no selection had been applied in this experiment, a genetic deterioration was not impossible. The experiment started with 24 families in 1906, 19 out of which either became naturally extinct or were disposed off before 1917, and only five families have continued throughout the 25 years period. If there were originally present some hereditary factors not favourable for vigour,or growth, the subsequent generations sooner or later would be affected by segregation.

Morris, Palmer and Kennedy (1933) seem to have been the first investigators to study the effect of selection and inbreeding on the efficiency of food utilization in albino rats. Two lines were selected from the F_2 generation of a cross between a pair of unrelated rats differed widely in their efficiency, one line from a high-efficient pair and the other from a low-efficient one. Between 500 to 600 individuals were observed to the F_9 generation. After the F_6 generation the average level of efficiency appeared uniform. The difference of efficiency between the two lines in the F_9 generation amounted to about 40 per cent. Their chief interest was to obtain a greater degree of uniformity in the utilization of food by rats, but they incidentally found that the correlation coefficient between dry matter consumed and gain in live weight for either sex was not high.

Winters and McMahon (1933) reported on their three series of experiments with 62 steers and found that the animals of the same breeding, age, weight, market grades and conditions exhibit significant differences in their abilities to make economical gains. The most significant indicators of net profit were daily rate of gain and final market evaluation.

Lush (1936) studied the genetic aspects of the swine progeny-testing in Denmark. During the years from 1920-1921 to 1934-1935 there were 7216 litters (each with four pigs) of Landrace breed pigs tested at five stations throughout the country. Between the years of 1922 and 1929 there was an improvement of 8 per cent in the efficiency of food utilization. In order to determine whether the improvement in the economy of gain was entirely a consequence of the more rapid growth or whether there was also some increase in physiological efficiency in addition to this, he computed the following figures, using the averages for the most dependable pre-war years, the three post-war years before the improvement began, and first five years after it ceased.

Period used	Average age at slaughter (days)	Total feed units used per unit of gain	Feed units per unit of gain, after deducting feed used for maintenance
1911-14	194.3	3.75	2.26
1920-23	196.0	3.63	2.10
1928-33	178.2	3.36	2.07

The table indicates that the improvement was mainly due to rapid gain along with a slight increase in physiological efficiency, which is shown in the last column. Further he believed that the faster growth was brought about by genetic changes in the swine population through selecting of breeding stock from more efficient animals. His belief is based on the two following facts: (1) the absence of recorded changes in feeds or management which might reasonably be supposed to have produced the improvement; (2) the correlation between relatives which show (especially for rate of gain) that there was considerable hereditary variability available for selection.

Burns (1937) of this Institute tested the efficiency of food utilization of four litters of rats totaling 34 animals, over a period of forty days and found definite differences between sexes and litters. She also stated that the growth rate was closely correlated with the efficiency of food utilization.

OBJECTS OF THE EXPERIMENT.

The objects of the investigation reported in this paper are outlined below:

(1) To determine the efficiency of food utilisation of albino rats reared in this laboratory.

(2) To study the relative importance of growth rate and food consumption in respect of efficiency of food utilization.

(3) To study the effect of seledtion and inbreeding on the efficiency of food utilization.in rats.

MATERIAL AND METHODS.

Animals.

Four litters of rats were chosen at random from the stock animals of this laboratory at the beginning of the experiment. No record was available of their pedigree or of the growth of their parents. These litters were designated as families A, B, C and D. Generations were indicated by a number affixed to the family, e.g. A_1 means the first generation put into this experiment. A total of one hundred and eleven rats have been tested. There were three generations in family A, with 18 rats; four generations in B, 29 rats; four generations in C, 26 rats; family D did not breed after the second generation, so there were only 8 individuals; four litters of controls, ($CK_{a, b, c, d}$) totaling 30 rats. The detail is given in Table 1.

Experimental period.

The animals were put under experiment for a period of eight weeks beginning at four weeks of age.

Management and feeding.

Reference to Part I, p. 20-22.

Breeding and selection.

After eight weeks' test the efficiency quotient of each member of the litter was calculated.

A male and a female, or a brother and sister, of the highest E.Q. value were selected for breeding. But in some cases, such as in A_2 generation there was only one female in the litter, therefore no selection was possible. The first litter from this couple were used for the same test when they were four weeks old, and the most efficient pair from this litter again sib-mated. The controls were chosen at random from stock rats.

Table 1

Number of Rats Used in the Experiment.

Family & generation	Males	Females	Total
A_1	2	4	6
A_2	3	1	4
A_3	2	6	8
B_1	2	2	4
B_2	1	4	5
B_3	7	3	10
B_4	8	2	10
C_1	2	2	4
C_2	2	4	6
C_3	3	3	6
C_4	4	6	10
D_1	1	5	6
D_2	1	1	2
CK_a	2	4	6
CK_b	4	4	8
CK_c	2	5	7
CK_d	3	6	9
Total	49	62	111

DISCUSSIONS OF EXPERIMENTAL RESULTS.

In Table 2 is given a summary of the observations made on gain in body weight, food consumption, and efficiency during the eight-week test feeding period, in terms of the means for each sex from the tested litters.

Sex difference in efficiency:- A comparison of the last two columns of the table shows a consistent difference between males and females similar to that previously observed. In general there is a tendency for both sexes to vary in the same direction. That is to say, when the males of the litter have a relatively low efficiency, the females of the same litter have also a relatively low efficiency. But the magnitude of the difference between the sexes seems to be affected by the level of the efficiency of the litter as a whole. Thus when the males are very efficient the sex difference is larger than when they are of low efficiency. For instance in litters A_1 and A_2 which are of low efficiency the sex difference is 0.054, and 0.076 in the value of the E.Q., but in litters B_3, B_4, and C_2, which are efficient litters, the difference amounts to 0.171,

Table 2

Efficiency Quotient, Gain in Body Weight and Food Consumption of Tested Rats.

Family & Generation	Efficiency Quotient				Average Gain (in grams)		Average Food Intake (in grams)	
	Average		Best pair					
	Male	Female	Male	Female	Male	Female	Male	Female
A_1	.154	.100	.155	.126	106	80	825	804
A_2	.196	.120	.209	.120	144	93	942	757
A_3	.248	.181	.271	.278	167	125	918	803
B_1	.255	.133	.261	.153	130	86	661	645
B_2	.228	.146	.228	.159	170	114	1083	887
B_3	.322	.151	.330	.172	200	120	972	873
B_4	.323	.163	.355	.172	191	121	887	863
C_1	.233	.112	.247	.125	141	93	777	788
C_2	.328	.157	.337	.185	207	124	1027	870
C_3	.240	.120	.252	.138	157	94	897	745
C_4	.256	.151	.287	.163	171	116	940	873
D_1	.074	.070	.074	.078	71	63	1007	884
D_2	.110	.123	.110	.123	85	100	598	812
CK_a	.197	.153	.208	.160	141	112	906	805
CK_b	.268	.151	.336	.160	161	104	897	770
CK_c	.255	.142	.278	.173	164	111	923	895
CK_d	.219	.145	.249	.158	140	103	834	746

0.160 and 0.171 respectively.

The lower efficiency of females has been discussed in several reports. Slonaker (1912) found that the female rat had a running activity of over 5000 miles during the whole life span, about two to four times greater than males. Mitchell, Card and Hamilton (1926) reported an analysis of the chemical composition of White Plymouth Rock chickens showing that the females had a higher percentage of dry matter and of ether extract than males. Mitchell and Hamilton (1927-1928) substantiated this finding by working on the chemical composition of beef cattle. Morris, Palmer and Kennedy (1933) analysed twelve male and twelve female rats and found the following results:

	Mean	
	Female	Male
Dry matter%	42.04±.42	37.50±.28
Nitrogen %	3.04±.03	3.28±.02
Protein %	18.97±.17	20.53±.09
Fat %	20.20±.56	14.11±.31
Fat-free dry matter %	21.77±.18	23.48±.09
Ash %	3.62±.04	3.25±.06
Final live weight, gm.	202.4 ±3.5	277.4 ±3.9
Final fat-free wt. gm.	161.6± 2.4	238.8 ±3.0
Total fat, gm.	40.2± 1.7	38.7 ±1.3

The female carcass had a higher energy value than the male carcass, having both higher dry matter and fat content. This would have to be taken into account in comparing efficiency of food utilization as between sexes.

Moreover, as has been demonstrated in Part I of this study the presence of the ovary hinders the body growth of females. Normal females of most species of animals grow a smaller size than males. Since the smaller the animal the greater relatively the surface area, females need relatively more energy for maintenance than males.

The relative importance of initial weight, gain in weight, and food consumption in determing the value of the E.Q.:- Some further examination of the factors influencing the value of the E.Q. has been attempted to see whether some information could be obtained about these differences. Since the E.Q. has been calculated from the formula:

$$E.Q. = \frac{\text{gain in weight}}{\text{food consumed}} \times \frac{(\text{initial wt.} + \frac{1}{2}\text{gain})}{100}$$

the E.Q, must obviously depend on each of the variables concerned. It is known however that the three variables are not necessarily uncorrelated themselves, so that change in E.Q. with variation in any one of the variables cannot be easily predicted. This difficulty has been overcome by calculating the standard partial regression coefficients in the way described by Snedecor (1938). These coefficients measure the relationship between the E.Q. and any of the three variables from which it is calculated independent of variation in the other two. Being in standard measure they are comparable with each other. The method of estimation is such that the variation in each variable is reduced to terms of its own standard deviation.

The first step was to calculate the total correlations linking the four variables. The values obtained are recorded in Table 3 for each sex separately.

Table 3

Correlation Coefficients.

	49 Male Rats		62 Female Rats	
r_{wg}	-0.164	NS	-0.376	SS
r_{wf}	-0.166	NS	+0.191	NS
r_{we}	-0.042	NS	-0.271	S
r_{gf}	+0.534	SS	+0.373	SS
r_{ge}	+0.923	SS	+0.942	SS
r_{fe}	+0.224	NS	+0.134	NS

w, initial weight in grams.
g, gain of body weight in grams.
f, total amount of food consumed.
e, efficiency quotient.

Among the males, the effect of initial weight on either gain, food consumption, or E.Q. appears to be negligible. In this sample of rats, the same does not apply to the females, among which the initial weight was negatively and significantly correlated with both gain and E.Q. Thus the higher the initial weight, the lower was the gain and the E.Q. Since E.Q. and gain in weight are strongly correlated, it may be suspected that influence of initial weight on E.Q. is derived from the fact that the higher the initial weight the sooner does the female reach the point when the growth rate diminishes and the E.Q. declines. Owing to the greater final weight of males, this consideration does not appear to have applied to the same extent. The positive correlation between gain in weight and food consumption agrees with the results of experiments elsewhere with rats, pigs and cattle, and is to be expected since the ratio of food energy available for growth to total energy of the food consumed increases with the growth rate. The size of the coefficients, however, indicates that a close linear relationship does not exist.

It is somewhat surprising that the correlation between food consumed and efficiency is not significant in either sex. As will now appear, however, this is due to the dependence of food consumption on initial weight and gain.

From these correlations, the standard partial regression coefficients given in Table 4 have been obtained.

Table 4.

Standard Partial Regression Coefficients.

	Males	Females
$\beta_{ew.gf}$	+ 0.101	+0.228
$\beta_{eg.wf}$	+ 1.139	+1.155
$\beta_{ef.gw}$	- 0.372	-0.340

Since all sources of variation in E.Q. have been accounted for all these regression coefficients are necessarily significant. But it is not to be concluded that the relationships indicated are true for all samples of rats.

It is clear that E.Q. is more strongly associated with gain in weight than with anything else. The faster the rats grew the more efficient they were. In comparison the influence of initial weight on E.Q. was small. In both sexes, there was a slight tendency for the rats which were initially heavier to be more efficient when differneces in gain and food consumption were eliminated. In both sexes also, the influence of initial weight was less than that of food consumed, although the latter too, appears to have been relatively unimportant. This is possibly due to the fact that the rate of food consumption did not alter very much with age. Since the rats were fed as much as they would eat, the negative relation between food consumption and E.Q. is interesting in view of the recent work on the restricted feeding of pigs (Mansfield et al, 1937). Among these rats, the elimination of effects of initial weight and gain, reveals the fact that those animals which ate less were more efficient than those which ate more than the average. The positive total correlations between food consumption and E.Q. are now seen to have been somewhat misleading.

The effects of selection and inbreeding;- Reference to Table 2 shows the progress which has been made during the short time available for selection for E.Q. In view of the results just discussed, it appears that selection for E.Q. will in practice have been selection for fast growth rate.

In families A and B, there is some evidence that the selection has been successful. In family C, no appreciable change can be seen. Animals belonging to family D were the worst of all the rats used in this experiment. There was only one male in the first generation. It put on only 71 grams of body weight but consumed 1007 grams of food, which is very seldom observed. The E.Q. of this rat was much lower than females of other families or controls. The five females of D_1 generation were as bad as the male. The pair selected from D_1 did not breed for a long time (about three months) then gave a litter of five, three of which died within the first two days after birth. The remaining male was a dwarf while very young, but during the experimental period it grew quite fast and had a 14 grams greater body weight increase than his father, with a decrease of food intake of 409 grams. The female gained 37 grams more and ate 72 grams less food than the D_1 females, so her E.Q. was higher.

The four litters of control animals are very uniform in growth of body weight and food consumption, and their E.Q. values may be classified as upper-middle class of this rat population.

Morris, Palmer and Kennedy (1933) have already obtained a high-level and a low-level strains of rats in food-utilizing efficiency, with a difference of 40% between the two. Our results are also supporting their findings. Lush (1936) has also shown that the improvement of efficiency of Danish swine population during the years 1922 to 1929 could be attributed to a great extent to the effect of selection of more efficient animals for breeding purpose, though not accompanied by inbreeding.

An extended discussion of the results so far obtained seems premature. Although selection within some of the families appears to have been successful, much caution needs to be exercised with such limited material. Large differences in E.Q. in the original parent pairs existed but it can not be assumed that all these were genetic in origin. In so far as they were caused by environmental differences, it would be expected that in some cases the offspring would show an improvement. There has also to be considered the possibility that with the passage of time, slight unintentional improvement in technique might occur which would have the appearance of genetic change. It is felt that many selection experiments have suffered in this way and that selection should be made to operate in both directions at once. If genetic differences exist in the original parents, it should be possible to create lines deviating in a positive as well as a negative way.

SUMMARY.

1. Inbreeding with selection for high Efficiency Quotient has been pratised on four families of rats. Together with their controls, a total of one hundred eleven rats have been individually fed for a period of eight weeks.

2. A close relation has been found to exist

between E.Q. and rate of gain. The influence of initial weight and food consumption has been much smaller. Similar results have been obtained from both sexes, and the indications are that rate of gain is an effective criterion by which to judge efficiency.

3. A clear sex difference in the level of food utilization has been observed. The lower efficiency of the females may be attributed to the energy value of their tissues, the inhibiting effect of ovary on growth, and also possibly to their greater spontaneous activity.

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